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Exosomes Mini-Series

A report on the International Society for Cell & Gene Therapy 2022 Scientific Signature Series, "Therapeutic advances with native and engineered human extracellular vesicles"

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ABSTRACT

The International Society for Cell & Gene Therapy Scientific Signature Series event "Therapeutic Advances With Native and Engineered Human EVs" took place as part of the International Society for Cell & Gene Therapy 2022 Annual Meeting, held from May 4 to 7, 2022, in San Francisco, California, USA. This was the first signature series event on extracellular vesicles (EVs) and a timely reflection of the growing interest in EVs, including both native and engineered human EVs, for therapeutic applications. The event successfully gathered academic and industrial key opinion leaders to discuss the current state of the art in developing and understanding native and engineered EVs and applying our knowledge toward advancing EV therapeutics. Latest advancements in understanding the mechanisms by which native and engineered EVs exert their therapeutic effects against different diseases in animal models were presented, with some diseases such as psoriasis and osteoarthritis already reaching clinical testing of EVs. The discussion also covered various aspects relevant to advancing the clinical translation of EV therapies, including EV preparation, manufacturing, consistency, site(s) of action, route(s) of administration, and luminal cargo delivery of RNA and other compounds.

Introduction

Extracellular vesicles (EVs) are a heterogeneous group of lipid membrane vesicles released by cells into the extracellular space. Although the term "exosomes" is widely acknowledged as a specific EV type derived from endosomes, in the industrial setting, it often is used to

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describe EVs that are smaller than 200 nm, termed small EVs (sEVs). Despite comparable sizes, sEVs comprise a very heterogenous population of EVs, with different functions and phenotypes. Regarding the functions of EVs, one early hypothesis was the disposal of cellular waste, where the EV collects and transfers waste to other cells such as the phagocytic cells or liver. However, the major role of EVs is now increasingly thought to mediate intercellular communication through the delivery of bioactive cargoes to elicit diverse biological responses in recipient cells. The bioactive cargo in EVs consists of a diverse array of molecules and is loaded by cells through specific processes before secretion. EVs then are able to deliver the cargo to specific cell and tissue types, where they elicit biological responses in recipient cells or tissues through cellular uptake, interaction with cellular receptors, or modulation of the microenvironment. As a result of the already-documented capacity for modification and efficient delivery, EVs are being considered for numerous therapeutic applications.

Therapeutic EVs can be broadly divided into native and engineered EVs. Native EVs refer to EVs secreted by cells where the EVs or secreting cells are not modified or engineered to modify the EV composition. Engineered EVs refer to those EVs modified to alter their composition after secretion or where cells are engineered to modify the EV composition.

Many cell types have been reported to secrete EVs that are therapeutically active. These include cell types such as mesenchymal stromal/stem cells (MSCs), neural stem cells (NSCs), endothelial cells, and immune cells, including macrophages. However, MSCs are currently the only cell type whose EVs have advanced to clinical trials. Of 11 clinical trials using native EVs, all were derived from MSCs (clinicaltrial.gov; search term: "exosomes," "extracellular vesicles" in Aug 2022) (Table 1). This could be attributed to the advanced clinical development of MSCs as therapeutic agents. Following hematopoietic stem cells, MSCs are the most clinically investigated cell type, with >1400 clinical trials registered at www.clinicaltrials.gov.

The possibility that EVs could be loaded with a specific payload where the payload is protected against inactivation and/or modified for delivery to specific target cells to elicit a desired response has made EVs tantalizing drug-delivery vehicles.

Sai Kiang Lim and Bernd Giebel organized, on behalf of the International Society for Cell & Gene Therapy (ISCT), a Scientific Signature Series event entitled "Therapeutic advances with native and engineered human EVs," as part of the ISCT 2022 meeting held from May 4 to 7, 2022, at the Moscone West Convention Centre in San Francisco, California, USA. This event was co-chaired by Daniel Weiss, former ISCT Chief Scientific Officer, and Eva Rohde, Chair of the International Society for Extracellular Vesicles Task Force on Regulatory Affairs and Clinical Use of EV-based Therapeutics. This event included presentations by invited speakers and discussions by panelists/attendees on how native and engineered human EVs are used in clinically relevant animal models or human trials. The discussion topics were wide-ranging and covered various aspects relevant to advancing the clinical translation of EV therapies, including EV preparation and consistency, the mechanism(s) of action, site(s) of action, route(s) of administration, and luminal cargo delivery of RNA and other compounds.

Table 1
Registered clinical trials using EVs and exosomes.

NCT number	Title
NCT05523011	Safety and Tolerability Study of MSC Exosome Ointment
NCT02138331	Effect of Microvesicles and Exosomes Therapy on β-cell Mass in Type I Diabetes Mellitus (T1DM)
NCT04798716	The Use of Exosomes for the Treatment of Acute Respiratory Distress Syndrome or Novel Coronavirus Pneumonia Caused by COVID-19
NCT05216562	Efficacy and Safety of EXOSOME-MSC Therapy to Reduce Hyper-inflammation In Moderate COVID-19 Patients
NCT03437759	MSC-Exos Promote Healing of Macular Holes
NCT04356300	Exosome of Mesenchymal Stem Cells for Multiple Organ Dysfunction Syndrome After Surgical Repair of Acute Type A Aortic Dissection
NCT04356300	A Pilot Clinical Study on Inhalation of Mesenchymal Stem Cells Exosomes Treating Severe Novel Coronavirus Pneumonia
NCT05060107	Intra-articular Injection of MSC-derived Exosomes in Knee Osteoarthritis (ExoOA-1)
NCT04388982	Safety and the Efficacy Evaluation of Allogenic Adipose MSC-Exos in Patients With Alzheimer's Disease
NCT04313647	AA Tolerance Clinical Study on Aerosol Inhalation of Mesenchymal Stem Cells Exosomes In Healthy Volunteers
NCT03384433	Allogenic Mesenchymal Stem Cell-Derived Exosome in Patients With Acute Ischemic Stroke

Native Human EVs

In the first half of the day at the Scientific Signature Series, speakers presented advances in therapeutic native EVs. As a reflection of the advanced state of clinical development of EV-based therapeutics, the first talk was the only one in this section that focused on another cell source other than MSCs. To this end, Randolph Corteling from ReNeuron started the Signature Series with his talk entitled "Neural Stem Cell-derived EVs as Novel Therapeutic Agents." He presented data on ReNeuron's conditionally immortalized clonally isolated human NSCs [1] that can be engineered to produce highly consistent EVs at a scale relevant for clinical development. He also shared that these cells can be engineered to carry a specific cargo for targeted delivery to specific regions in the brain and that EVs from different human NSC lines derived from specific brain regions (cortex, striatum, hippocampus, and ventral mesencephalon) have discrete surface profiles that confer distinct EV tropism for specific regions in the brain. To further influence the EV tropism, he also reported that after intrathecal delivery, human NSCs being engineered to express brain-derived neurotrophic factor on the EV surface home to the striatum and hippocampus, in contrast to their native counterparts. In addition to human NSC lines, ReNeuron also obtained immortalized cell lines derived from the human liver, retinal and pancreatic progenitors, and inducible pluripotent stem cells (iPSCs) for EV production.

In contrast to NSC and other cell-derived EVs, native MSC-EV products are in an advanced state of clinical development. Still, their use as therapeutic agents remains challenging, especially when primary MSCs are used as the cellular source of EVs. As summarized by Bernd Giebel in the second talk of this session on "Functional Heterogeneity Among Independent MSC-EV Preparations," primary MSCs are highly heterogenous, and many factors contribute to this heterogeneity, such as tissue of origin, differences in donor profile, isolation method, and culture system. He showed that functional MSC heterogeneity is inherited in their EV products. Using a graft-versus-host disease mouse model, he demonstrated heterogeneities caused by variable donors and intra-donor heterogeneities within the expanded MSC populations. Routinely, MSCs are raised and expanded as oligoclonal cell stocks. During MSC expansion, the oligoclonal cell stocks seem to undergo random clonal selection [2], resulting in unavoidable batch-to-batch variations in MSC and MSC-EV products. Accordingly, Bernd Giebel considered that primary MSC products might never reach market readiness due to the different heterogeneity levels. To overcome this and other limitations, his group adopted a previous concept to mitigate the impact of MSC heterogeneity on EV products by immortalizing primary MSCs and establishing monoclonal MSC lines [3]. He showed that EV preparations from the derived monoclonal MSC lines exhibit comparable *in vitro* and *in vivo* activities in modulating proinflammatory reactions to those from their primary counterparts. His group's next goal is to set up and optimize a scaled, Good Manufacturing Practice–compliant MSC-EV production process. Considering that production process parameters affect the potency of resulting EV products (i.e., the process defines the product), he also briefly elaborated on the requirement of appropriate

quality control assays, including potency testing for the product development of functionally active MSC-EV products and their release into clinical testing [4–6].

In the next talk, "A Potency Assay for MSC-EV in Reducing IL-17 in Psoriasis," Sai Kiang Lim further elaborated on the importance of potency testing in developing MSC-EV drug products. She emphasized the elucidation of the mechanism in developing a potency assay using the topical use of MSC-EVs to treat psoriasis as an example. Psoriasis is a common inflammatory skin condition that affects approximately 1 in 100 of the population and is characterized by the rapid multiplication of skin cells leading to skin thickening and scaling. Using EVs from an immortalized MSC line [3], she described the mechanism of action by which her topically applied MSC-EV product reduces the level of interleukin (IL)-17 and C5b-9 terminal complement complex in a mouse model of psoriasis [7]. First, the site of action was determined to be the stratum corneum, as topically applied MSC-EV product was found to be physically confined to the stratum corneum of human skin explants. Because psoriasis is characterized by the accumulation of neutrophils known as Munro's microabscesses and activated complements in the stratum corneum, the likely targets for the topically applied MSC-EV product were neutrophils and complements. Using human neutrophils and complements, it was demonstrated that MSC-EVs could significantly reduce the formation of C5b-9, which in turn reduce complement-induced neutrophil extracellular trap release and IL-17 production. Her team further revealed that the MSC-EV product inhibits complement activation through CD59 [8]. On the basis of these findings, CD59 is the critical MSC-EV attribute in mediating MSC-EV activity against psoriasis. The level of CD59 in the MSC-EV preparation is thus predictive of the MSC-EV potency against psoriasis. Her team has recently completed a Phase I clinical trial of 10 patients (ClinicalTrials.gov ID: NCT05523011) and reported no adverse reactions.

The following talk also focused on using an MSC-EV product in a dermatological condition. Byong Seung Cho, in his talk "Potential Clinical Applications of Adipose Stem Cell-Derived sEVs for the Treatment of Atopic Dermatitis and COVID-19," described the therapeutic efficacy of human adipose MSC-EVs in alleviating atopic dermatitis. Atopic dermatitis is a chronic relapsing inflammatory skin condition. It is the most common skin disease in children, affecting approximately 15–20% of children and 1–3% of adults. In mouse models of atopic dermatitis, subcutaneous injections of human adipose MSC-EVs reduced local inflammation and infiltration of mast cells and inflammatory dendritic epidermal cells; suppressed systemic inflammation by decreasing the levels of serum IgE, eosinophils, and cytokines such as IL-4, IL-31, tumor necrosis factor (TNF)-α and IL-23 as well as promoted epidermal barrier repair by inducing *de novo* synthesis of ceramides, an effect that was not observed with dexamethasone treatment [9,10]. He also presented the anti-viral activity of human adipose MSC-EVs. In a mouse model infected with the H1N1 influenza A virus, treatment with human adipose MSC-EVs enhanced the percentage survival rate, clinical score, body weight, and temperature. Histological analysis of the lungs further showed improved histopathological scores and reduced levels of inflammatory cytokines such as IL-1β, IL-6, IFN-γ, and TNF-α. This anti-viral activity could potentially be applied against coronavirus disease 2019.

Next, Josephine Herz, in her talk "Impact of MSC-EVs on Neonatal Hypoxic–Ischemic Brain Injury," reported on the impact of EV products obtained from primary as well as clonally expanded immortalized MSCs on modulating the symptoms of neonatal brain injury caused by hypoxia–ischemia (HI). HI is a leading cause of childhood mortality and neurodevelopmental morbidity, which involves a pronounced neuroinflammatory response. Josephine Herz demonstrated that intraperitoneal injections of MSC-EVs were significantly protective from HI-induced striatal tissue loss by mounting a multi-faceted response of reducing neuroinflammation, promoting neural proliferation, and improving oligodendrocyte maturation [11]. Addressing the MSC heterogeneity issue, her team has further demonstrated in the same model that clonally expanded immortalized MSCs MSC-EVs provided by the group of Bernd Giebel exerted comparable neuroprotective effects as EVs from the original primary MSCs.

Showing the clinical potential of MSC-EV products, Mario Gimona, in his talk "MSC-EV for the Improvement of Intracochlear Implant Function," reported the results of MSC-EV application in cochlea implantation. Cochlea implantation is a conventional therapy for people with hearing loss, a common problem among people older than 65 years of age. To aid in hearing, cochlear electrode arrays frequently are implanted, which in a proportion of patients induce fibrotic capsule formation, significantly impeding the electrical performance of the cochlear implant. Upon co-applying umbilical cord-MSC-EVs with a cochlear implant, the survival and neurite extension of spiral ganglion neurons were increased. Upfront, the applied MSC-EV product inhibited mitogen-activated and phytohemagglutinin-induced T-cell proliferation and alleviated lipopolysaccharide-induced inflammation of microglial cells [12]. The local application of MSC-EVs to the inner ear attenuated hearing loss and protected auditory hair cells in a noise trauma mouse model [12]. These pre-clinical findings provided the scientific rationale for clinical testing. In an individual treatment performed on a 55-year-old patient with Ménière's disease, the team intraoperatively applied allogeneic umbilical cord-MSC-EVs before inserting a cochlear implant [13]. Safety and efficacy evaluation after 24 months showed no increase in electrical impedance and no adverse effects [13].

Another clinical application of an MSC-EV product was reported by Maroun Khoury in his talk "Phase I Clinical Trial: MSC-EVs for the Treatment of Knee Osteoarthritis." Osteoarthritis (OA), the most common joint disorder, affecting more than 300 million individuals worldwide, has an increasing prevalence due to aging and obesity [14]. He presented data showing the positive effects of intra-articular injections of human umbilical cord–derived MSC-EVs on joint repair and pain recovery in a mouse model of knee OA. A bio-distribution study further demonstrated no EV release or leakage from the injection site for up to 72 hours after injection. miRNA profiling of the EVs identified miRNAs involved in regulating the genes and factors involved in cartilage destruction. Next, he presented the case study findings of a 56-year-old female patient with symptomatic knee OA. Remarkably, the patient showed improvements in clinical scores after EV treatment. For future work, after clinical-grade characterization and preparation, it is planned that a single dose of MSC-EVs will be administered to 10 patients with moderate knee OA through intra-articular injection in a registered clinical trial (ClinicalTrials.gov ID: NCT05060107).

The next talk also focused on using an MSC-EV product for joint repair. In his talk "MSC-EV Therapy for Joint Injuries and Osteoarthritis," Wei Seong Toh reported that weekly intra-articular injections of MSC-EVs enhanced cartilage regeneration and alleviated pain and degeneration in rat models of the osteochondral defect and OA [15,16]. Unlike MSC-EVs derived from primary cell and tissue sources, the MSC-EVs were derived from the immortalized human MSC line of Sai Kiang Lim's group, which ensues an infinite supply of cells for scalable production of MSC-EVs [3]. Enhanced cartilage regeneration was attributed to chondrocyte migration, proliferation, and matrix synthesis, mediated partly through CD73-mediated adenosine activation of protein kinase B (AKT), extracellular signal-regulated kinase (ERK), and adenosine monophosphate-activated protein kinase (AMPK) signaling pathways. On the basis of these findings, CD73 is a critical MSC-EV attribute mediating MSC-EVs' effects in cartilage repair. Therefore, the measurement of CD73 activity could be useful in predicting the potency of MSC-EV preparations for cartilage repair. As a prelude to a clinical trial, his team recently demonstrated the safety and efficacy of human MSC-EVs for cartilage repair in a clinically relevant large animal model [17]. They showed that the combination of MSC-EVs and hyaluronic acid administered at a clinically

acceptable frequency of three intra-articular injections at weekly intervals could promote functional cartilage and subchondral bone repair, with significantly improved morphological, histological, and mechanical outcomes in a clinically relevant porcine model. Importantly, no adverse response was observed in any of the animals. The study highlights a clinically translatable protocol using human MSC-EVs to treat patients with cartilage lesions and potentially OA.

MSC-EV products are not only considered a potential drug for degenerative diseases but may also impact the aging processes. Aging is characterized by the progressive decline in homeostatic and regenerative capacities, partly attributed to the loss of functional resident stem cells necessary for maintaining tissue homeostasis. Paul Robbins, in his talk "MSC-EV as Anti-Aging Therapeutics," presented data on muscle-derived stem/progenitor cells (MDSPCs) that are adversely affected by aging. Specifically, his group demonstrated that MDSPCs isolated from old and ERCC1-deficient progeroid mice are defective in proliferation and multi-lineage differentiation, and this stem cell dysfunction directly contributes to age-related degeneration [18]. Transplantation of young MDSPCs was able to extend the health span and life span of ERCC1-deficient mice. However, the transplanted MDSPCs did not differentiate or migrate from the injection site, suggesting that the therapeutic effects of MDSPCs were mediated by secreted factors [18]. His team further demonstrated that EVs derived from bone marrow MSCs of young mice and from the immortalized human MSC line of Sai Kiang Lim's group had senotherapeutic capacity to attenuate senescence in cultured senescent fibroblasts and in naturally aged mice and in ERCC1-deficient mice, improving measures of healthspan *in vivo* [19]. His results identified EVs as key factors released by young MSCs that can reduce cellular senescence, rescue stem cell dysfunction, and extend health span.

Altogether, the presentations on native MSC-EV products demonstrate that despite being produced by different MSC sources using various enrichment protocols, they could improve disease symptoms across multiple disease models. However, it is widely recognized that clinical translation of these experimental observations will be critically dependent on the development of rigorous potency assays to ensure that the therapeutic potency of the EV product is reproducible from the laboratory to clinical testing and finally to the market [5,6].

Engineered Human EVs

In the second half of the session, speakers presented advances in engineered EVs for therapeutic applications. The session began with Samir El Andaloussi's talk on "EV Engineering for Directed Loading and Delivery of Biotherapeutics." He described strategies for identifying the best exosomal sorting domains for cytoplasmic delivery. His team combined the coding regions of 250 different proteins, including tetraspanins, PTGFRN, and gag (a viral protein), that had been annotated to sort into EVs with different reporter proteins of different sizes. More than 20 of them robustly allowed luminal EV loading across different cell types. To investigate the functional delivery capacity of different scaffold proteins, CRE recombinase was used as the payload. More than 50 different engineered EV types were tested in different loxP reporter cells. Fusion constructs that promoted the greatest CRE loading also conferred the greatest rate of recombination in reporter cells. To robustly induce recombination in recipient cells, the inclusion of fusogenic proteins is recommended due to the limited endosomal escape of EVs. Upon administration of CRE-loaded EVs into floxed reporter mice, recombination was observed in different organs. Interestingly, even EVs devoid of a fusogenic protein promoted CRE-mediated recombination to a certain degree, at least when injected locally, implying that luminal cargo delivery *in vivo* is different from *in vitro*. It needs more research to unravel the mechanisms of how endosomal escape is mediated in these cells. Similarly, his group demonstrated that by using the same setup but linking an RNA binding domain to EV scaffolds and co-expressing such fusion protein with shRNA, robust silencing was observed in recipient cells. In addition to this, Andaloussi also touched upon the extensive work they have conducted on studying the pharmacokinetics and pharmacodynamics of injected EVs in mice. Using bioluminescence imaging, they have shown that systemically injected EVs distribute rapidly body-wide to all organs, but that signals also decline rapidly. The half-life of most tested EVs is short, but by engineering the second loop of tetraspanins with albumin binding domains, the circulation time of EVs is significantly enhanced [20].

Chulhee Choi from Ilias Biologics presented his talk entitled "Intracellular Delivery of NF-κB Inhibitor Via Optogenetic Engineering of Extracellular Vesicles, From Bench to Clinic." He reported the development of EXPLOR (Exosome engineering for protein loading via optically reversible protein-protein interactions), an optogenetic system in which two protein domains originally found in plants reversibly aggregate upon blue light illumination. Upon fusing one of the domains to CD9 and the other to a protein normally not delivered into EVs, proteins of interest can be loaded into EVs. [21]. Using this technology, Ilias Biologics engineered HEK293T cell-derived exosomes to contain a non-degradable form of IκB (super-repressor lκB, or srlκB), a protein that binds to nuclear factor-κB and prevents its translocation into nuclei and thus activation of proinflammatory programs. The therapeutic efficacy of such engineered exosomes (Exo-srlκB) was demonstrated in mouse models of sepsis [22] and kidney ischemia–reperfusion injury [23]. In septic mouse models, intraperitoneal injection of Exo-srlκB alleviated kidney injury, systemic inflammation and improved overall survival [22]. Similarly, in a mouse model of renal ischemia–reperfusion injury, Exo-srlκB treatment downregulated NF-κB signaling and ameliorated inflammation and apoptosis in ischemic kidney injury [23].

Douglas Williams from Codiak presented his talk on "Therapeutic EVs: Engineering, Manufacturing, and Clinical Activity." He described the company's engEx toolkit for therapeutic exosome development, including engineering and production, to overcome the scalability and delivery issues. He also emphasized the importance of bioreactors for continued high viability and purity products while eliminating scale-up risk. Furthermore, he introduced five exosome-based products in the oncology platform, including candidates to circumvent pharmacokinetics/pharmacodynamics issues of STING agonism, solving long-standing issues of IL-12 biology in cutaneous T-cell lymphoma, enabling targeted nucleic acid delivery to myeloid-rich cancers and inhibiting tumor growth in the B16F10 Luc tumor. He described the "Catch AND Release" platform on the gene delivery platform where EVs were used to deliver the AAV gene. Functional AAV capsids remain inside EVs by BASP-1 (for luminal loading) with 2-3 log improvement of loading. Using EVs for vaccination is also in the pipeline of Codiak for novel vaccines aiming to produce robust mucosal CD8 T cell responses and deliver antigen(s), adjuvant, and co-stimulators to the antigen-presenting cells.

In the last session, Tony De Fougerolles from Evox Therapeutics presented "Engineered Exosomes: Therapeutic Advances and Platform." He described the company's proprietary DeliverEX platform for engineering the exosomes to carry diverse drug cargoes and designing them to target different tissues. In an example, he presented data on the surface engineering of exosomes to display tumor necrosis factor receptor 1 and interleukin-6 signal transducer as decoy receptors for TNF-α and IL-6, respectively [24]. By genetic engineering of the exosome-producing cells to express oligomerized exosomal sorting domains and the N-terminal fragment of syntenin, the efficiency and inhibitory activity of tumor necrosis factor receptor 1 and interleukin-6 signal transducer and their display on the exosomes were enhanced. In mouse models of systemic inflammation, neuroinflammation, and intestinal inflammation, exosomes displaying the cytokine decoys ameliorated the disease phenotypes with greater efficacy than clinically used agents targeting the TNF-α and IL-6 pathways [24]. Next, he presented preliminary data on exosome

delivery of soluble argininosuccinate lyase (ASL) via intraluminal loading for the potential treatment of Argininosuccinic aciduria, a rare genetic disorder characterized by the deficiency of the enzyme ASL, resulting in elevated levels of ammonia in the blood. He showed that treating exosomes containing human ASL could reduce the plasma ammonia levels in ASL-deficient mice.

This session presented a spectrum of creative tools to engineer EVs for specific therapeutic functions. The speakers also highlighted a need to consider different routes of administration to ensure delivery of the engineered EVs to specific cell types and to further engineer EVs to define their subsequent fate after cellular uptake.

Conclusions

The meeting concluded with the appreciation that despite significant progress made in several aspects relevant to the clinical translation of EV therapies, there are still many unresolved EV issues, such as biodistribution, pharmacodynamics, and cellular fate after uptake. The importance of these unresolved issues in the clinical translation of EVs is moderated by the route of EV administration and disease target. For example, these issues are mitigated in the topical application of exosomes where the distribution of EVs is limited to the non-living stratum corneum. In contrast, these issues would be important when EVs were injected intraperitoneally or intratumorally. Engineered EVs designed to deliver cargo intracellularly will have to be evaluated for cellular uptake and post-uptake fate.

Declaration of Competing Interest

WST is a scientific advisory board member of Paracrine Therapeutics. SKL is the founder of Paracrine Therapeutics and has a scientific advisory role at Ilias Biologics, and ExoCoBio Inc. BG is a scientific advisory board member of Innovex Therapeutics SL and Mursla Ltd, a consultant of Fujifilm and a founding director of Exosla Ltd. MK is the chief scientific officer of Cells for Cells and REGENERO. PDR is on the scientific advisory board for Unicyte Gmbh. RC is an employee of ReNeuron Limited.

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Author Contributions

Conception and design of the study: BG and SKL. Acquisition of data: WST, RY, SEIA, BSC, CC, RC, ADF, MG, JH, MK, PDR, DW, DJW and ER. Analysis and interpretation of data: WST, RY, SEIA, BSC, CC, RC, ADF, MG, JH, MK, PDR, DW, DJW and ER. Drafting or revising the manuscript: WST, RY, BG and SKL. All authors have approved the final article.

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