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Review

Extracellular vesicles derived from mesenchymal stem cells: the wine in Hebe's hands to treat skin aging

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Abstract

Owing to its constant exposure to the external environment and various stimuli, skin ranks among the organs most vulnerable to manifestations of aging. Preventing and delaying skin aging has become one of the prominent research subjects in recent years. Mesenchymal stem cells (MSCs) are multipotent stem cells derived from mesoderm with high self-renewal ability and multilineage differentiation potential. MSC-derived extracellular vesicles (MSC-EVs) are nanoscale biological vesicles that facilitate intercellular communication and regulate biological behavior. Recent studies have shown that MSC-EVs have potential applications in anti-aging therapy due to their anti-inflammatory, anti-oxidative stress, and wound healing promoting abilities. This review presents the latest progress of MSC-EVs in delaying skin aging. It mainly includes the MSC-EVs promoting the proliferation and migration of keratinocytes and fibroblasts, reducing the expression of matrix metalloproteinases, resisting oxidative stress, and regulating inflammation. We then briefly discuss the recently discovered treatment methods of MSC-EVs in the field of skin anti-aging. Moreover, the advantages and limitations of EV-based treatments are also presented.

Keywords: mesenchymal stem cell; extracellular vesicles; exosomes; skin aging; anti-aging; photoaging

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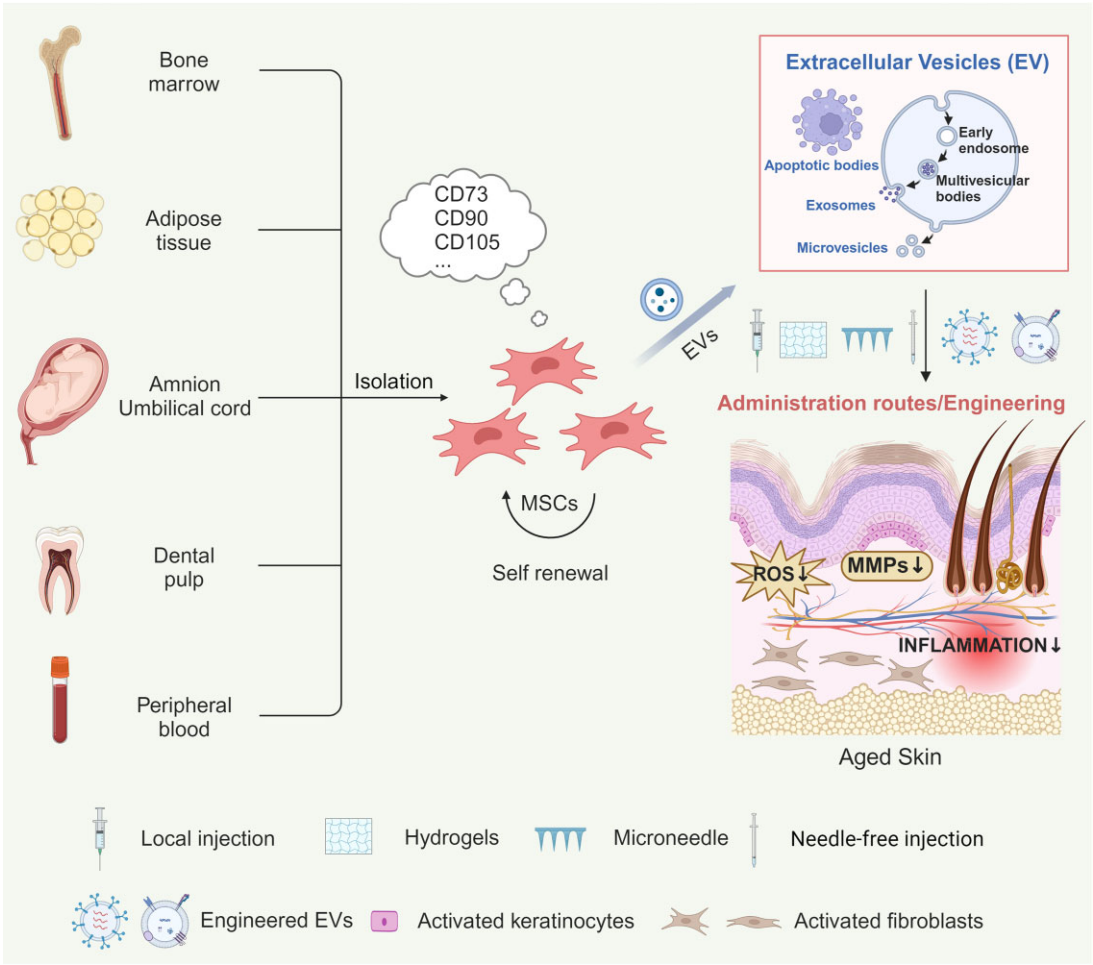
Graphical Abstract



Introduction

As the largest organ of the human body, the skin not only serves the purpose of external beauty but also plays multiple crucial physiological and biological roles. It acts as the first line of defense for our body, playing a key role in preventing the invasion of harmful substances and microorganisms from the external environment [1]. Due to continuous exposure to environmental factors, skin aging begins from birth. It is a multifactorial process associated with tissue and organ degeneration, increasing the risk of mortality [2]. Skin aging is a complex and multifaceted phenomenon that has been extensively studied in the fields of dermatology, biology, and cosmetic science. This natural process is influenced by both internal and external factors, which collectively contribute to the appearance of visible signs of aging in the skin [3]. Understanding the mechanisms that lead to skin aging is crucial for developing effective preventive measures and treatment methods.

Many components have been proven to potentially help delay skin aging. For example, antioxidants such as vitamin C, vitamin E, and coenzyme Q (CoQ10) can protect the skin from damage caused by free radicals, exhibiting antioxidative and anti-inflammatory effects [4–6]. Collagen peptides contribute to improving skin elasticity [7]. Hyaluronic acid (HA) helps reduce skin moisture loss [8]. Green tea extract, rich in antioxidants, possesses anti-aging properties [9]. Despite the increasing number of researchers involved in studying the delay of skin aging, current treatment methods have not yet achieved ideal results, necessitating the exploration of more effective treatment strategies.

Mesenchymal stem cells (MSCs) are a type of multipotent stromal cell with self-renewal and multi-lineage differentiation capabilities. They originate from the mesodermal tissue and are widely distributed in the connective tissues and stromal compartments of various organs throughout the body [10]. Under specific *in vivo* or *in vitro* induction conditions, MSCs have the potential to differentiate into various tissue cells, including adipocytes, osteocytes, chondrocytes, myocytes, tenocytes, ligamentocytes, neurons, hepatocytes, cardiomyocytes, and endothelial cells [11]. This multidirectional differentiation potential makes MSCs highly promising in the fields of tissue engineering, regenerative medicine, and clinical therapy, as they can be used to repair or replace damaged tissues, offering new hope for the treatment of many diseases. In addition to their multi-lineage differentiation potential, MSCs can also secrete soluble factors, including cytokines, chemokines, and growth factors [10]. Over the past few decades, MSCs have gained widespread attention and research as an effective cellular therapy in various medical fields. However, some people believe that stem cell therapy has certain drawbacks and limitations, including limited differentiation potential, risk of immune rejection, ethical and moral concerns, and lack of sufficient clinical evidence to support its application. Recent studies have found that extracellular vesicles (EVs) produced by MSCs may play a crucial role in the therapeutic effects of MSC treatment. This discovery provides

the possibility for the development of a new cell-free treatment approach [12].

EVs are nanosized vesicles with phospholipid bilayer membranes [13]. Based on their diameter, EVs can be categorized into six subpopulations: exomeres (<50 nm), exosomes (30–150 nm), ectosomes or shedding microvesicles (100–1000 nm), apoptotic bodies (1000–5000 nm), migrasomes (500–3000 nm), and large oncosomes (1000–10 000 nm). Exosomes are currently a highly researched subtype in the field of EVs due to their small size and various functions within the organism. Early studies in the 1980s initially characterized exosomes as endosomal vesicles secreted by reticulocytes. For a substantial period, exosomes were regarded as cellular repositories for "debris" or "garbage bags". Nevertheless, over the past decade, there has been a renewed surge in interest in EVs due to their remarkable role in intercellular communication and immune response. EVs, enveloped by lipid bilayers, harbor a multitude of molecular components, including nucleic acids, proteins, and lipids. These molecules, such as RNAs and proteins, are selectively packaged into cargo and delivered to target cells through receptor-mediated mechanisms [14]. Leveraging this mechanism, researchers have employed bioengineering techniques to engineer EVs, facilitating the loading of synthetic drug molecules, thereby enhancing drug delivery efficiency and target specificity [15]. Therefore, EVs may be a potentially innovative therapeutic strategy that promises to play a key role in the field of skin anti-aging. In this comprehensive review, we provide an overview of the molecular mechanisms underlying skin aging and discuss the applications and therapeutic avenues as well as the advantages and limitations of MSC-EVs in delaying skin aging.

Mechanisms of skin aging

The skin serves as a protective barrier, separating our internal organs from the external environment. It plays a crucial role in defending against pathogenic invasions and safeguarding us from physical and chemical harm [16]. Epidermis, dermis, and subcutaneous tissue (hypodermis), are the three layers of the skin, each with different cell types. The epidermis, which forms the outermost layer of the skin and covers an average area of 2 m^2, is primarily composed of keratinocytes. It also contains Langerhans cells (LCs), melanocytes, and Merkel cells. These cells form a physical barrier through tight junctions and desmosomes to prevent water loss and microbial invasion. Beneath the epidermis lies the dermal layer, which includes a multitude of fibroblasts, immune cells, blood vessels, sweat glands, hair follicles, and nerve fibers. Effective communication between these various cell types and the extracellular matrix (ECM) is crucial for maintaining tissue integrity. The subcutaneous tissue, the deepest layer of the skin, consists of adipose tissue, typically originating from loose connective tissue. Adipose tissue plays a significant role in mechanical protection, insulation, and energy storage [17, 18].

The skin is capable of maintaining internal balance and responding to external challenges through effective intercellular communication. Various signaling mechanisms, such as paracrine and autocrine signaling, enable cells to convey information about changes in the microenvironment, regulating physiological processes like inflammation, wound healing, cell proliferation, and immune responses. When the skin is exposed to various external stimuli, including ultraviolet (UV) radiation, pollutants, and pathogens, these stimuli can generate reactive oxygen species (ROS) and trigger inflammatory responses. ROS are necessary for cellular signaling but can be damaging in excess. Skin cells employ complex signaling pathways to balance the accumulation of ROS, including antioxidants and repair mechanisms. Similarly, the inflammatory response is coordinated through a series of events involving various cell types, including dendritic cells, macrophages, and neutrophils. This complex network, composed of different cell types and signaling pathways, ensures that the skin can rapidly adapt to changes in the environment [19–21].

The process of skin aging is complex, inevitable, and multifaceted [22]. Skin aging is primarily divided into intrinsic aging and extrinsic aging, with the former occurring with the passage of time and the latter being caused by external factors such as UV radiation and environmental pollutants [23]. Below, we will further discuss the pathological and physiological mechanisms underlying these two types of aging.

Pathophysiological mechanisms in intrinsic and extrinsic aging

Intrinsic aging represents a physiological process primarily linked to genetic, hormonal, and metabolic factors [24]. Clinical manifestations include skin thinning, dryness, reduced elasticity, the appearance of fine lines, and impaired skin repair [25]. As individuals age, certain genes within senescent cells undergo alterations, including genes responsible for encoding interleukins and matrix metalloproteinases (MMPs). These genes, known as senescence genes, can impact cell function, repair mechanisms, and overall physiological processes, thereby playing a role in the aging process [26, 27]. Intrinsic aging also involves alterations in skin structure and reduced sensitivity of fibroblasts to mechanical stimulation, leading to decreased dermal collagen synthesis and an increase in collagen bundle thickness, which, in turn, delays the skin wound healing process [28]. Additionally, epidermal thinning results in compromised skin barrier function, characterized by increased moisture loss from the epidermis due to keratinocyte atrophy [29]. Furthermore, intrinsic skin aging has been associated with endocrine dysregulation, such as decreased estrogen levels post-menopause [30, 31].

Extrinsic aging is primarily instigated by lifestyle choices and exposure to external environmental factors such as UV radiation, air pollution, and temperature fluctuations [32]. Photoaging refers to the skin alterations induced by exposure to UV radiation, accounting for ~80% of facial aging. Its primary characteristics include a reduction in skin epidermal thickness, pigmentation deposition, the development of wrinkles, skin laxity, erythema, and an increased risk of malignancy [33]. UV radiation is categorized into three subtypes based on wavelength ranges: UVA (320–400 nm), UVB (280–320 nm), and UVC (200–280 nm) [22]. UVA radiation comprises the predominant portion of the sun's UV rays, possessing significant penetrating capability. It can penetrate deep into the dermal layer, extending beyond the epidermis, where it causes damage to dermal fibroblasts and reduces collagen production, ultimately disrupting the skin's normal structure [34]. UVB radiation indeed possesses greater energy than UVA, and only a fraction of UVB rays can penetrate the ozone layer to reach the Earth's surface [35]. UVB radiation primarily affects the keratinocytes of the epidermis. The high energy levels associated with UVB radiation can directly damage DNA by inducing double-strand breaks and causing the formation of thymine-thymine cyclobutane pyrimidine dimers. These DNA alterations have the potential to contribute to the development of certain skin cancers and premature skin aging [36]. As for UVC radiation, it is the most potent type of UV radiation. However, it is completely absorbed and filtered by the ozone layer, preventing it from reaching the Earth's surface (Fig. 1) [37].

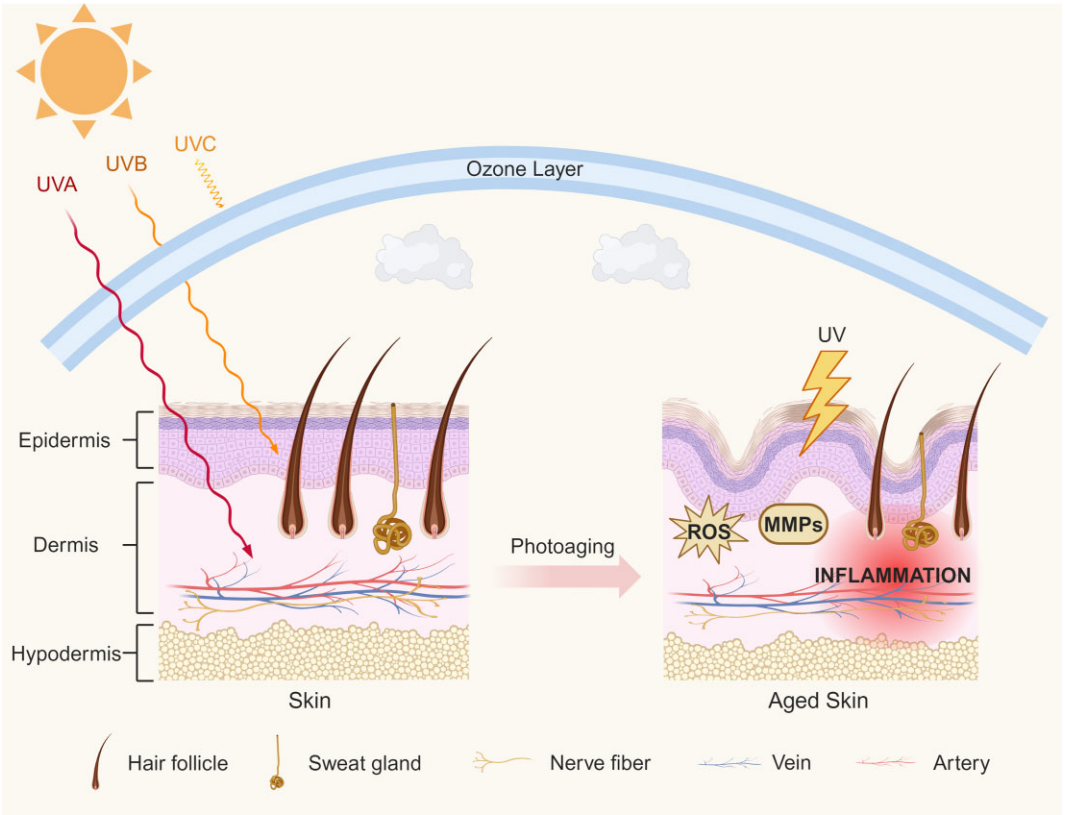


Figure 1. Mechanisms of skin photoaging caused by UV radiation. The skin can be divided into three layers: the epidermis, the dermis, and the hypodermis. UVA has the longest wavelength and can penetrate into the dermis layer of the skin. UVB has a shorter wavelength than UVA and can penetrate into the epidermis layer of the skin, but a significant portion of UVB is absorbed by the ozone layer. UVC has the shortest wavelength and is entirely absorbed by the ozone layer. UV radiation causes an increase in the production of ROS and MMPs in the skin as well as an inflammatory response.

Molecular mechanisms in skin aging

Oxidative stress

Oxidative stress constitutes a shared factor associated with both intrinsic and extrinsic aging processes. Elevated levels of ROS contribute to oxidative stress, characterized by an imbalance favoring prooxidants over antioxidants [38]. Excessive oxidative stress has been linked to skin cancers, including malignant melanoma [39]. ROS exert regulatory control over the activation of MMPs, which are crucial in the remodeling of the ECM and are believed to play a pivotal role in the pathogenesis of skin aging. Notable members of the MMP family involved in this process include MMP-1, MMP-2, MMP-3, and MMP-9 [40]. ROS function as second messengers by activating transcription factors, such as nuclear factor-kappa B (NF-κB) and activating protein-1 (AP-1). NF-κB, in addition to up-regulating MMP-1, MMP-3, and MMP-9, leading to the degradation of various collagen fibers, also promotes the expression of pro-inflammatory cytokines, such as tumor necrosis factor-α (TNF-α) and interleukin-6 (IL-6). AP-1, comprising c-Fos and c-Jun subunits, operates downstream of the activation of mitogen-activated protein kinases (MAPKs) [41]. The three principal members within the MAPK family encompass the extracellular signal-regulated kinase (ERK), the C-Jun N-terminal kinases (JNKs), and the p38 MAPKs. Upon ROS-induced activation of MAPK signaling cascades, phosphorylation of ERK and JNK stimulates c-Fos and c-Jun, culminating in the increased expression of the MMP family. Additionally, AP-1 modulates the TGF-beta/Smad pathway by inhibiting the expression of the Smad2/3 complex, leading to a reduction in type I procollagen synthesis (Fig. 2) [42–44].

Telomere shortening

Telomeres are specialized nucleoprotein structures located at the terminal ends of eukaryotic chromosomes. They consist of tandem double-stranded hexanucleotide repeats (TTAGGG) that serve as protective caps, shielding chromosome ends from degradation, end-to-end fusion, and homologous recombination. Due to the inability of DNA polymerases to fully replicate chromosome ends, telomeres naturally shorten by ~50–200 bp with each cell division. When telomeres reach a critical length, DNA ends become exposed, triggering the DNA damage response and ultimately leading to cellular senescence [45]. Telomerase, a ribonucleoprotein complex, is comprised of telomerase reverse transcriptase and RNA-dependent DNA polymerase. Telomerase can utilize its RNA component as a template to elongate the telomere sequence, thus preserving telomere integrity and stability [46]. Several studies have proposed a potential link between telomeres, telomerase, and skin aging. Victorelli *et al.* [47] reported that senescent epidermal melanocytes exhibit dysfunctional telomeres, inducing telomeric damage in peripheral keratinocytes through paracrine signaling, thereby contributing to skin aging. Jia *et al.* [48] observed significant telomere shortening in human skin fibroblasts following exposure to UVA radiation. Marion *et al.* [49] found that telomeres in dermal skin fibroblasts from young mice (22 weeks old) were longer than those from aged mice (121 weeks old). Additionally, Flores *et al.* [50] reported that telomerase-deficient mice displayed a propensity for premature skin aging. However, the direct causal relationship between telomeres, telomerase, and skin aging remains to be definitively established. Consequently, further

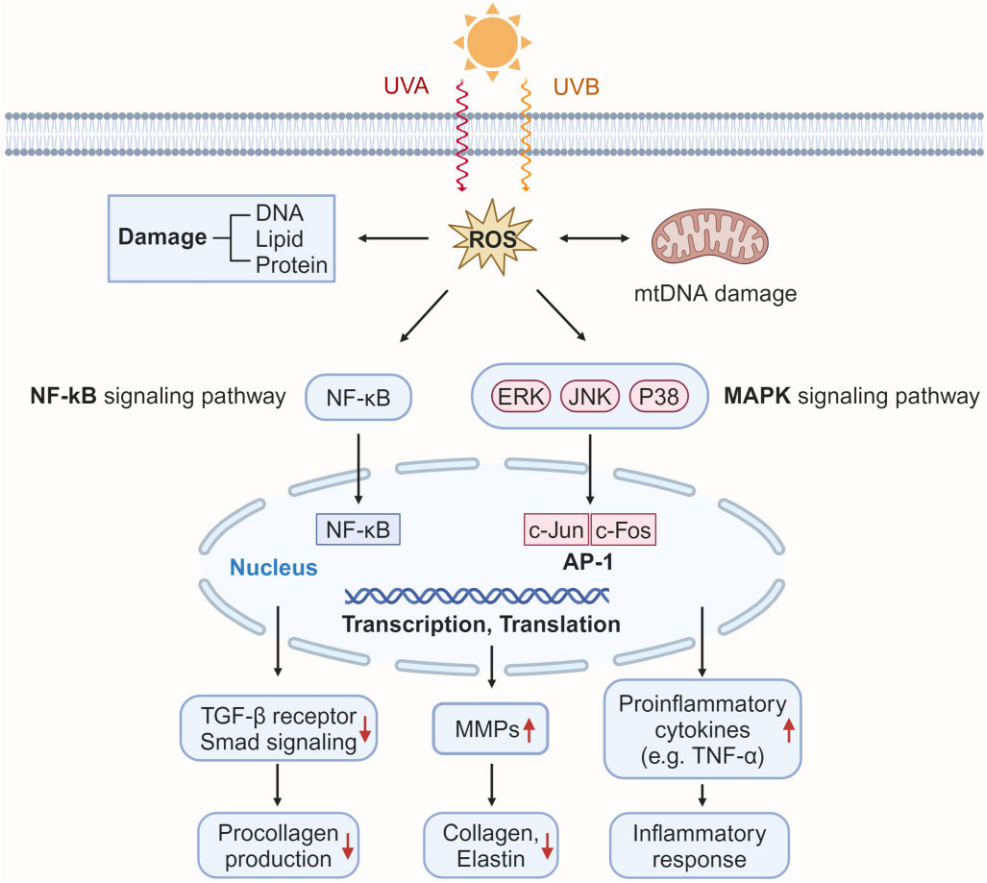


Figure 2. Mechanisms of skin photoaging mediated by UV via ROS.

comprehensive investigations are warranted to delve deeper into the functions of telomeres and telomerase in the context of cellular senescence and skin aging.

Mitochondria and melatonin

Within eukaryotic cells, mitochondria play a pivotal role in energy production [51]. The skin, a highly organized organ constantly renewing itself throughout life, relies on the proliferation, differentiation, and self-renewal of stem cells for regeneration and repair [52]. These stem cells meet their energy requirements through oxidative phosphorylation, a process in which mitochondria generate ATP. However, a consequence of mitochondrial respiration is the continuous production of ROS. Mitochondrial DNA (mtDNA) is particularly vulnerable to oxidative stress-induced damage due to its close proximity to ROS production sites. Prolonged UV radiation causes a rise in ROS in skin cells as well as mtDNA damage, which may disrupt normal mitochondrial function and intracellular energy production [53–55]. Berneburg *et al.* [56] reported that 2 weeks of UVA radiation increased human skin mtDNA deletions. Schroeder *et al.* [57] found that human skin fibroblasts with partial depletion of mtDNA exhibited a photoaging-like phenotype as evidenced by decreased mitochondrial function, increased MMP-1 expression, and decreased matrix alpha1 type-I collagen (COL1A1) expression. Furthermore, there is an important antioxidant and ROS scavenger, coenzyme Q (CoQ10), present in the mitochondria, which is believed to possess anti-aging activity [58]. The synthesis of CoQ10 was observed to decrease with aging [59]. Marcheggiani *et al.* [60] reported that the reduction of CoQ10 affects the respiratory efficiency of mitochondria, leading to oxidative damage and mitochondrial dysfunction, ultimately inducing premature aging of human dermal fibroblasts (HDFs).

Melatonin, a natural antioxidant primarily synthesized by the pineal gland and released into the bloodstream to regulate circadian rhythms, is also produced by the skin and the gastrointestinal tract [61]. Dermal fibroblasts express melatonin receptor MT-1. Dong *et al.* [62] have provided evidence that the MT-1 receptor may play a role in protecting skin cells from UV-induced DNA damage. They observed that MT-1 receptor expression in fibroblasts decreased with age. The use of small interfering RNA technology to knock down the MT-1 receptor resulted in fibroblasts that were more susceptible to UV irradiation, exhibiting a significant increase in oxidative stress and DNA damage. Moreover, a dynamic melatonin–mitochondria axis exists within the skin. On one hand, melatonin maintains mitochondrial homeostasis to preserve cellular redox balance. Kleszczyński *et al.* [63] found that melatonin prevents dissipation of mitochondrial transmembrane potential after UV radiation, thereby reducing apoptosis. Mansouri *et al.* [64] reported that melatonin attenuated ethanol-induced deletion of mtDNA. Díaz-Casado *et al.* [65] found that melatonin restored ATP production and the respiratory capacity of dysfunctional mitochondria and reduced oxidative stress. On the other hand, mitochondria contain high levels of melatonin and serve as the site of melatonin synthesis and metabolism in skin cells [66–68]. Two melatonin metabolic pathways, the mitochondrial cytochrome P450-dependent pathway and the kynuric pathway, are found in mitochondria. Melatonin synthesis and metabolism are closely related to mitochondrial function [69]. The interactions

between melatonin and mitochondria have garnered increasing attention from researchers.

The role of microRNAs

MicroRNAs (miRNAs) are short noncoding RNA molecules, typically consisting of 19 to 25 nucleotides, which regulate gene expression post-transcriptionally by interacting with target messenger RNAs (mRNAs) [70]. Several miRNAs have emerged as key regulatory molecules implicated in skin aging processes. Ahmed *et al.* [71] identified an increase in miR-21 expression in both mouse and human aged skin. miRNA-21 (miR-21) plays a role in altering the expression of age-related genes (Krt1, Krt10, and Krt17) by targeting the down-regulation of the chromatin remodeler SATB1 in keratinocytes. This alteration promotes skin aging and heightens susceptibility to age-related pathological conditions. Srivastava *et al.* [72] reported significant changes in miRNA expression in photoaged skin. Solar exposure leads to the upregulation of miR-34a, miR-134, miR-145, and miR-383, alongside the downregulation of miR-663b, miR-3648, and miR-6879. Among these, miR-34a, miR-134, and miR-383 may play roles in both intrinsic and extrinsic aging. Furthermore, Rock *et al.* [73] discovered increased expression of miR-23a-3p and reduced secretion of HA in senescent dermal fibroblasts. MiR-23a-3p acts as a regulator by binding to the 3' untranslated region of hyaluronan synthase 2, resulting in decreased HA synthesis, promoting cellular senescence and contributing to skin aging. While numerous studies have highlighted the importance of miRNAs in the processes of chronological aging and photoaging of the skin, the specific underlying mechanisms and pathways remain to be fully elucidated. Further research is needed to explore and investigate these mechanisms comprehensively.

Inflammaging and immunosenescence

Inflammation is a hallmark feature of photoaging, where repeated exposure to UV radiation triggers heightened oxidative stress and localized skin inflammation. This oxidative stress and inflammation lead to cellular senescence, and the buildup of senescent cells acts as a catalyst for increased production of inflammatory cytokines. This shift in the balance of pro-inflammatory and anti-inflammatory factors results in a state of chronic low-grade inflammation known as "inflammaging". Inflammaging does not just affect the skin, it can also contribute to systemic diseases such as Parkinson's disease, cardiovascular disease, type 2 diabetes, and cancer. Chronic low-grade inflammation can increase susceptibility to infections and trigger age-related immune deficiencies, collectively referred to as immunosenescence. Recent evidence suggests that both inflammaging and immunosenescence play significant roles in skin aging [74–76]. LCs, specialized antigen-presenting cells in the epidermis, are affected by aging. Pilkington *et al.* [77] reported that the number of LCs in older skin is significantly reduced, by ~20%, compared to younger skin. Furthermore, the migration ability of these cells is impaired, which increases the susceptibility of elderly individuals to skin infections. Macrophages, innate immune cells, are crucial players in inflammation. Macrophages can adopt two main phenotypes: "pro-inflammatory" M1 and "anti-inflammatory" M2 [78]. Gather *et al.* [79] found that the inflammaging microenvironment in aging skin promotes the differentiation of monocytes into macrophages with "pro-inflammatory" M1-like characteristics, resulting in an increased number of M1 macrophages. Conversely, senescent monocytes exhibit reduced intrinsic differentiation into M1 macrophages. This shift toward more M1 macrophages in the skin contributes to ECM remodeling and decreases the overall stability of skin tissue. While studies have underscored the pivotal roles of inflammation and immunity in the accumulation of physiological and pathological damage during aging, the precise molecular mechanisms involved remain poorly understood. Further investigations are warranted in the future to shed more light on these mechanisms and their impact on skin aging.

Autophagy

Autophagy is a crucial lysosome-dependent degradation pathway in eukaryotic cells that plays a pivotal role in maintaining intracellular homeostasis [80]. Dysregulated autophagy has been implicated in various diseases, including skin aging, atherosclerosis, and cancer [81]. In the context of UV-induced skin photodamage, autophagy regulation is vital for cellular protection. Umar *et al.* [82] demonstrated that intervening in autophagy significantly attenuated UVB radiation-induced nuclear damage and apoptosis in HDFs. However, UVB exposure led to an impaired autophagic response in HDFs. Treatment with the autophagy activator rapamycin improved autophagy levels by reducing oxidative stress and promoting DNA repair, ultimately reducing the number of apoptotic cells. Several studies have reported that modulating autophagy with certain compounds can slow down the skin aging process. For instance, metformin, an oral hypoglycemic drug commonly used to treat type II diabetes, has been found to mitigate skin photoaging. Chen *et al.* [83] reported that metformin exerts its anti-photoaging effects by inhibiting mitochondrial autophagy, reducing oxidative stress, and decreasing the activation of the PI3K/AKT/mTOR signaling pathway in senescent cells. *In vivo* experiments confirmed that metformin treatment significantly reduced collagen breakdown and epidermal thinning in UVA-induced skin damage in mice, thereby improving photoaged skin conditions. Caffeine, a potential neuroprotective agent with antioxidant effects [84], has also been shown to activate autophagy and reduce UV-induced skin damage in mice. Li *et al.* [85] found that low doses of caffeine activate autophagy through the A2AR/SIRT3/AMPK pathway, reducing the effects of oxidative stress on epidermal keratinocytes. Notably, the use of autophagy inhibitors diminished the protective effect of caffeine. While both the activation and inhibition of autophagy have been linked to skin aging, the precise molecular mechanisms remain incompletely understood. Further research is needed to solidify the evidence and gain a deeper understanding of these mechanisms in the context of skin aging.

Applications of MSC-derived EVs against skin aging

MSCs are multipotent stem cells capable of self-renewal and differentiation into various cell types, both mesenchymal and non-mesenchymal [86]. They can be derived from diverse sources, including bone marrow (BMSCs), adipose tissue (ADSCs), umbilical cords (uMSCs), dental tissue (dental pulp stem cells), and peripheral blood [87]. MSCs typically express common surface markers like CD73, CD90, and CD105, while lacking hematopoietic markers like CD14, CD19, CD34, CD45, CD11b, CD79alpha, and HLA-DR [88].

Nearly all cells including eukaryotes and prokaryotes have been demonstrated to release membrane-enclosed vesicles called EVs. Exosomes are a subclass of EVs, with a diameter of 40–160 nm [89]. Trams *et al.* [90] in 1981 first proposed the term "exosome" as 5′-nucleotidase activity microvesicles released by different cultured cells. Over an extended period of time, exosomes

were considered as cellular "garbage dumpsters". However, extensive research has since illuminated their crucial role in intercellular communication and signal transduction. Exosomes are capable of carrying various cargoes, including proteins, lipids, and nucleic acids, making them instrumental in cell-to-cell communication [91]. Fusion of multivesicular bodies with the plasma membrane secretes exosomes into the extracellular environment [92]. Once released by parent cells, exosomes can be taken up by recipient cells through various mechanisms such as endocytosis, membrane fusion, or receptor–ligand interactions. Importantly, exosomes released by different types or functional states of cells exhibit heterogeneity. The efficiency of exosome internalization may depend on the nature of the cargo contained within these vesicles and the metabolic state of the recipient cells. Due to their excellent biocompatibility and low biotoxicity, as well as their pivotal role in cellular communication, exosomes are emerging as ideal candidates for gene and drug delivery [89, 93, 94]. For instance, miR-21, an oncogenic miRNA, is found to be overexpressed in various malignancies like lung cancer, breast cancer, and colon cancer. Downregulating miR-21 has been shown to inhibit the proliferation of tumor cells, promote apoptosis, and enhance tumor sensitivity to chemotherapy. Liang *et al*. [95] significantly improved the efficacy of 5-FU-resistant colon cancer treatment by encapsulating chemotherapy drugs like 5-FU and miR-21 inhibitor oligonucleotides within engineered exosomes. This innovative approach holds great potential in targeted drug delivery. As a result, there is a growing interest in leveraging the advantages of exosomes as novel nano-drug delivery vehicles, leading to extensive research into the specific mechanisms of targeted drug delivery. This area of study has the potential to revolutionize drug delivery methods, making them more precise and efficient.

MSCs have emerged as prolific producers of therapeutic extracellular vesicles, making them a valuable resource in various medical applications [96]. Compared with other therapeutic programs, the advantages of MSC-EVs in the treatment of skin aging are focused on the following aspects. The first is security: as EVs are vesicles, they avoid risks associated with the administration of living cells [97, 98], such as microvascular obstruction [99], infusion toxicity, and ectopic tissue formation [100]. This is in contrast to nano-cosmeceuticals, in which active ingredients are packaged within a nanocarrier so that they are easily absorbed into the skin and subsequently exert enhanced cosmetic and therapeutic effects to improve skin aging [101]. The most commonly used nanocarriers in the cosmeceutical field include liposomes, vesicles, solid lipid nanoparticles (NPs), niosomes, nanocapsules, micelles, dendrimers, and metal NPs [102, 103]. However, some of these NPs may produce toxicity or unwanted side effects such as activation of the innate immune system, inflammation, and skin irritation due to their composition, particle size, and electrical charge [104]. MSC-EVs completely avoid the adverse effects of nano-cosmeceuticals and their composition is closer to nature. The second is high efficiency: as MSC-EVs are derived from MSCs and are one of the main ways of intercellular communication [105]. The nucleic acid and other contents loaded by MSC-EVs regulate the biological time or the change of cell fate, which is an extremely efficient mode of action [106, 107]. It induces a more efficient and comprehensive change than cosmetics, supplemental HA or collagen. The third is targeting and tissue regeneration: EVs can specifically target cells through their surface proteins, promoting skin regeneration and repair. This targeted delivery provides potential advantages over traditional treatment methods, allowing for more accurate action on damaged skin tissue [108, 109]. The final is feasibility: with the deepening of research, the storage problem of EVs has also been solved to a certain extent [110, 111], which provides a high possibility of future commercial applications.

In summary, MSC-EVs offer exciting prospects for addressing skin aging and promoting skin rejuvenation. A deeper understanding of the specific mechanisms underlying EV-mediated effects in skin aging is anticipated to provide valuable insights into these processes and guide further advancements in the field of skin rejuvenation and tissue regeneration.

MSC-EVs on epidermal cells and fibroblasts

The epidermis comprises four distinct layers: the outermost stratum corneum, followed by the stratum granulosum, stratum spinosum, and stratum basale. Sustained proliferation and differentiation of keratinocytes, originating from the basal layer at the dermal–epidermal junction, contribute to the formation of the stratum corneum, a vital component in maintaining the dynamic homeostasis of the epidermis [112]. Located beneath the epidermis, the dermis serves as its foundational support and is connected to the epidermis through the dermal–epidermal junction. The dermis primarily consists of fibroblasts and their ECM, encompassing collagen, elastin, and various proteins responsible for the skin's mechanical strength and elasticity [113]. With advancing age, both the epidermis and dermis gradually thin, accompanied by a decline in the self-renewal capacity of skin stem cells, ultimately leading to compromised skin barrier function. Single-cell transcriptome profiles of human eyelid skin across different age groups have indicated a decline in the overall population of keratinocytes and fibroblasts among the elderly compared to their younger counterparts. In fibroblasts, the inactivation of HES1 (growth-controlling transcription factors) and in keratinocytes, the inactivation of KLF6 (a crucial regulatory factor in the transcription process of epidermal basal cells) leads to cell aging. The data suggests that, for keratinocytes, the downregulation of KLF6 not only results in decreased proliferative potential but also leads to heightened expression of pro-inflammatory cytokines, including IL-6. Concerning fibroblasts, exposure to UV radiation triggers the downregulation of the Notch-targeting gene HES1. Conversely, the overexpression of the HES1 gene has shown promise in mitigating age-related phenotypes. These findings offer novel insights into potential therapeutic targets for addressing skin aging and related conditions [114]. Several studies have underscored the role of MSCs in promoting the healing of damaged skin tissue through paracrine mechanisms. Chang *et al*. [115] have demonstrated that the overexpression of the long non-coding RNA FOXD2-AS1 in exosomes derived from ADSCs (ADSC-Exos) accelerates the migration and proliferation of HaCaT cells by influencing the miR-185–5p/ROCK2 axis, thereby facilitating wound healing. Furthermore, Zhao *et al*. [116] have observed that human uMSC-derived exosomes (HuMSC-Exos) effectively inhibit H_2O_2-induced apoptosis in HaCaT cells and promote tissue regeneration in skin wounds. This was established using a HaCaT cell-based skin injury model treated with H_2O_2. Results indicated that HuMSC-Exos significantly curtailed the nuclear translocation of apoptosis-inducing factor (AIF) and the excessive activation of poly ADP ribose polymerase 1 (PARP-1), thus mitigating HaCaT apoptosis attributed to decreased mitochondrial membrane permeability. Additionally, *in vivo* administration of HuMSC-Exos upregulated endothelial cell markers CD31 and cytokeratin 10 while downregulating the expression of alpha-smooth muscle actin (alpha-SMA). Zhang *et al*. [117] have reported that ADSC-Exos expedite the healing of full-thickness skin wounds by

stimulating HDF proliferation and migration, as well as fostering the synthesis of type I and type III collagen through the PI3K/Akt signaling pathway. The TGF-beta1/Smad signaling pathway, which plays a pivotal role in the formation of collagen and elastin fibers during wound healing, has also been implicated. Jiang *et al*. [118] have highlighted that human BMSC-derived exosomes (BMSC-Exos) expedite the wound healing process by inhibiting the TGF-beta/Smad signaling pathway. *In vitro*, exosomes have been shown to enhance the proliferation of HaCaT and HDF. *In vivo* studies have demonstrated that exosomes downregulate the expression of TGF-beta1, Smad2, Smad3, and Smad4, while upregulating the expression of TGF-beta3 and Smad7, thereby promoting scar-free wound healing. Notably, exosomes exhibit more significant therapeutic effects compared to BMSCs in this context. These findings hold promise for advancing the application of cell-free therapy in the future.

Removal of MMPs

MMPs are zinc-dependent neutral endopeptidases intricately involved in the remodeling of the ECM. The protein hydrolytic activities of MMPs undergo regulation through transcription, proenzyme activation, and interactions with tissue inhibitors of metalloproteinase (TIMPs). The role of TIMPs lies in their control of ECM degradation by binding to activated MMPs. An MMP/TIMP ratio imbalance has been linked to various diseases, including atherosclerosis, osteoarthritis, and cancer [119]. In the context of skin, prolonged exposure to UV radiation induces an upsurge in MMPs production within fibroblasts and keratinocytes, resulting in the degradation of normal collagen and elastin fibers, ultimately leading to skin damage and the potential development of skin cancer [120]. Notably, senescent fibroblasts can induce collagen degradation through the secretion of senescence-associated secretory phenotype (SASP), mediating MMP-1 expression in normal fibroblasts. SASP also induces dysfunction and premature senescence in adjacent cells, propagating senescence [121]. Previous investigations have demonstrated that UV irradiation activates the MAPK and NF-κB signaling pathways, leading to an upregulation in the production of MMP-1, MMP-2, MMP-3, and MMP-9 [40, 122]. Currently, numerous studies have posited that MSC-derived exosomes (MSC-Exos) play a pivotal role in suppressing MMP expression. Choi *et al*. [23] have demonstrated that EVs derived from ADSCs (ADSC-EVs) restore the migration and proliferation of UVB-damaged HDFs. This restorative effect is attributed to the ability of ADSC-EVs to stimulate dermal matrix synthesis by promoting the expression of TIMP-1 and TGF-beta1, inhibiting the overexpression of MMP-1, -2, -3, and -9, and enhancing the expression of collagen types I, II, III, and V and elastin. Deng *et al*. [123] reported that HuMSC-derived extracellular vesicles (HuMSC-EVs) shield HDFs from UVB-induced cell death and growth arrest, simultaneously reducing the percentage of senescent cells and MMP-1 expression while promoting Col-1 expression. Notably, miR-1246 was found to be downregulated during the photoaging process of HDFs. Gao *et al*. [124] conducted experiments wherein ADSCs were infected with lentivirus to obtain miR-1246-overexpressing exosomes, exploring their effects on HDFs after UVB irradiation. *In vitro*, miR-1246-overexpressing exosomes attenuate MMP-1 expression and enhance the secretion of type I procollagen by inhibiting the MAPK/AP-1 signaling pathway and activating the TGF-beta/Smad pathway, respectively. *In vivo*, miR-1246-overexpressing exosomes mitigate epidermal thickness, wrinkles, and collagen loss in UVB-induced mice.

Protection against oxidative stress

UV radiation can induce the mitochondria-dependent generation of ROS, which can inflict oxidative damage upon DNA bases [125]. ROS, including superoxide anions, hydroxyl radicals, and H_2O_2, serve as essential players in cell signaling and the maintenance of cellular homeostasis. At low concentrations, ROS function as signaling molecules, governing processes such as cell proliferation, differentiation, growth, and survival. However, when present in high concentrations, ROS trigger oxidative stress damage, culminating in cell necrosis and apoptosis [126]. The cellular ROS level is intricately tied to the regulation of collagen metabolism; elevated ROS levels activate MMP-1, MMP-3, and MMP-9, initiating collagen degradation and, consequently, affecting the assembly and turnover of the ECM [127]. Under normal physiological conditions, the antioxidant defense system, composed of glutathione peroxidase (GPx), superoxide dismutase (SOD), peroxidase (POD), and catalase (CAT), is activated to control ROS accumulation, thereby averting excessive ROS production, which could otherwise lead to extensive lipid peroxidation damaging cell membranes and causing cell death. Nevertheless, during oxidative stress, these antioxidant systems may prove insufficient to scavenge excess ROS [128, 129]. Emerging evidence suggests that MSC-Exos can bolster cellular antioxidant capabilities and modulate ROS levels to mitigate oxidative damage. Wang *et al*. [130] reported that MSC-Exos can repair skin damage induced by oxidative stress, involving the NRF2 signaling pathway. *In vitro* results revealed that MSC-Exos reversed abnormal calcium oscillations and mitochondrial alterations in H_2O_2-exposed keratinocytes, concurrently elevating levels of ferric reducing antioxidant power (FRAP), fGPx, and SOD to curtail ROS production. *In vivo*, MSC-Exos counteracted the increased number of mitochondrial cristae and swelling observed in UV-irradiated mouse skin, while also reducing the expression of CAT, SOD2, GLUT1, and pro-inflammatory cytokines like TNFalpha, IL-1beta, and IL-6, and enhancing COL1 and COL3 deposition. MSC-Exos exert their antioxidative prowess both *in vitro* and *in vivo* by adaptively regulating the NRF2 defense system. Shiekh *et al*. [131] produced an elastic antioxidant polyurethane biomaterial (OxOBand) combined with ADSC-Exos. They found that OxOBand was able to reduce oxidative stress and increase collagen remodeling to promote diabetic wound healing. Li *et al*. [132] found that the overexpression of Nrf2 in ADSC-Exos accelerates diabetic wound healing by reducing ROS levels and inhibiting the expression of inflammatory factors. Wu *et al*. [133] reported that HuMSC-Exos-delivered 14–3-3ζ protein promoted the expression of SIRT1 in HaCaTs, which significantly reduced the elevation of ROS caused by UV radiation.

Regulation of inflammation

Inflammation constitutes a pivotal factor in the process of cellular senescence and ranks among the primary characteristics of skin photoaging. As individuals age, the features of inflammation closely parallel changes in immune cell function, a phenomenon termed immunosenescence [134,135]. With age, the innate immune system experiences a decline in the ability of dendritic cells to activate $CD4^+$ T cells. Concurrently, the adaptive immune system undergoes thymic degeneration, leading to reduced output of total naïve T cells and an increased proportion of memory T cells [136, 137]. This age-associated immunosenescence, both innate and adaptive, coincides with chronic, low-grade systemic inflammation, aptly termed inflammaging [138]. Continuous exposure of the skin to UV radiation induces localized inflammation and a counterproductive immunosuppressive

milieu. Hasegawa *et al*. [139] have demonstrated that UVB radiation-induced DNA damage activates the NLRP3 inflammasome, resulting in the release of inflammatory mediators such as IL-1beta, PGE2, TNF-alpha, IL-1alpha, and IL-6. CCAAT/enhancer-binding protein beta (C/EBPbeta), a transcription factor regulating the expression of the IL-1beta gene, plays a pivotal role in this process. Xiao *et al*. [140] exposed keratinocytes to 50 mJ/cm^2 UVB radiation to create an *in vitro* model of acute injury. UVB radiation induced the secretion of various pro-inflammatory cytokines, including IL-1beta, TNF-alpha, and FGF-2, while upregulating the expression of C/EBPbeta and promoting nuclear translocation. Fortunately, MSCs possess potent anti-inflammatory and immunomodulatory properties, as evidenced in various models of inflammatory diseases, including arthritis, psoriasis, and systemic lupus erythematosus [141]. Psoriasis, a chronic inflammatory skin disorder with genetic and environmental factors at its core, exemplifies this. Zhang *et al*. [142] reported that the topical application of MSC-Exos reduced the release of psoriasis-specific inflammatory cytokines IL-17 and IL-23, along with the membrane attack complex C5b-9, in a mouse model of psoriasis. Cho *et al*. [143] demonstrated that ADSCs-Exos ameliorated symptoms of atopic dermatitis by reducing the number of CD86$^+$ and CD206$^+$ cells, eosinophils, and mast cells, while inhibiting the expression of inflammatory factors TNF-alpha, IL-23, IL-31, and IL-4 in an atopic dermatitis mouse model. In terms of skin regeneration, many wounds stall in the healing process due to persistent inflammation, ultimately developing into chronic, non-healing wounds. These recalcitrant wounds, if infected and left unaddressed, can lead to severe complications, including amputation and even mortality. Patel *et al*. [144] reported that long non-coding RNA GAS5 in ADSCs-Exos reduces levels of IFNalpha, IL-1beta, and TNF-alpha by suppressing toll-like receptor 7 expression in HDFs, thereby expediting wound healing in a chronically inflamed environment. Liu *et al*. [145] observed that melatonin-stimulated exosomes from human BMSCs significantly enhanced the healing of diabetic wounds. These exosomes shifted pro-inflammatory M1 macrophages towards an anti-inflammatory M2 phenotype through regulation of the PI3K/AKT signaling pathway, thereby ameliorating the inflammatory status of the wound and facilitating healing. Heo [146] explored the impact of pretreating BMSCs with selenium and collecting their exosomes on wound healing. Their results demonstrated that selenium-primed BMSCs exhibited heightened anti-inflammatory capabilities. Selenium-boosted exosomes inhibited TNF-alpha, IL-6, and IL-8 while promoting the expression of TGF-beta1 and IL-10 in monocytes. Additionally, selenium-boosted exosomes facilitated angiogenesis and regulated the inflammatory microenvironment by promoting M1-to-M2 macrophage polarization. Overall, selenium-boosted exosomes promoted wound healing by dampening inflammation and modulating angiogenesis (Fig. 3, Table 1).

Clinical study of MSC-EVs in the treatment of aging skin

To date, few clinical studies have explored the potential of MSC-EVs for therapeutic effects on skin aging, although encouraging findings have been reported in many *in vitro* and preclinical studies. We checked clinicaltrials.gov and have not found any of the above studies ongoing. Two clinical trials of high relevance to the field of skin therapy are underway. One of the clinical trials (No. PTD2021P001) is exploring the safety of topical MSC-derived exosome ointment. Another clinical trial (No. AGLE-102–102) is to explore the safety and efficacy of allogeneic MSC-EVs in deep second-degree burn wounds. However, as of this writing, no follow-up reports have been seen on these two clinical trials.

In a recently reported 12-week clinical study, researchers evaluated the use of microneedles (MN) combined with ADSC-Exos to treat skin aging, using a prospective, randomized, splitface, comparative study. A total of 28 individuals received three treatments, with an interval of 3 weeks, and were followed up for 6 weeks after the last treatment. The results showed that compared with the control group of MN combined with saline, the overall aesthetic improvement scale score of the experimental group was significantly higher than that of the control group [147].

Administration routes and engineering of MSC-EVs

Despite the notable effect of MSC-EVs in the treatment of photoaging, how to administer the drug in the case of the skin, which is the natural barrier of the organism, has also become a topic of concern for scholars. In addition, as research has progressed, many modifications have been employed to optimize the targeting and efficiency of EV treatments to more fully realize the potential of MSC-EVs. Here, we summarize the common methods of drug delivery, as well as the modification of EVs by engineering methods, and the treatment of photoaging in combination with other therapeutic approaches.

Local injection

Appropriate administration methods can effectively improve the uptake rate of EVs, so as to achieve the desired therapeutic effect. Compared with systemic administration, topical administration has a unique attraction, such as direct delivery of high concentrations of EVs to the injured site, improving the ability of recipient cells to absorb EVs and reducing the probability of side effects. In general, systemic administration generally requires a higher total dose per patient than local administration [148]. EVs are rapidly cleared following systemic administration [149]. In the process of clinical application of drugs, efficiency, simplicity, and cost are important factors that must be considered when choosing the route of administration. Since skin photoaging problems are usually concentrated in exposed skin sites, such as faces and hands, localized administration is an effective approach to address this issue. Compared with direct application, the injection method can directly deliver the drug to the target site, thereby bypassing the skin barrier. Therefore, local injection has become the mode of administration in most studies.

Hydrogels

Hydrogels are 3D nanofiber materials composed of physically or chemically cross-linked hydrophilic polymer networks. They possess unique properties, such as the ability to absorb and retain a significant amount of water while maintaining their structural integrity and dimensions [150]. Hydrogels are ideal therapeutic platforms due to their biocompatibility and modifiability, as appropriate functional groups can be added as needed.

The use of hydrogels has been widely explored in many biomedical fields, including cell therapy, drug delivery, biosensing, and tissue engineering [151–153]. Encapsulating EVs in a hydrogel can maintain their activity while exerting a sustained release effect. Mol *et al*. have developed a hydrogel known as UPy-hydrogel, which is based on ureido-pyrimidinone (UPy) units coupled to poly (ethylene glycol) chains [154]. This hydrogel has been investigated as a potential delivery platform for EVs. The UPy-hydrogel

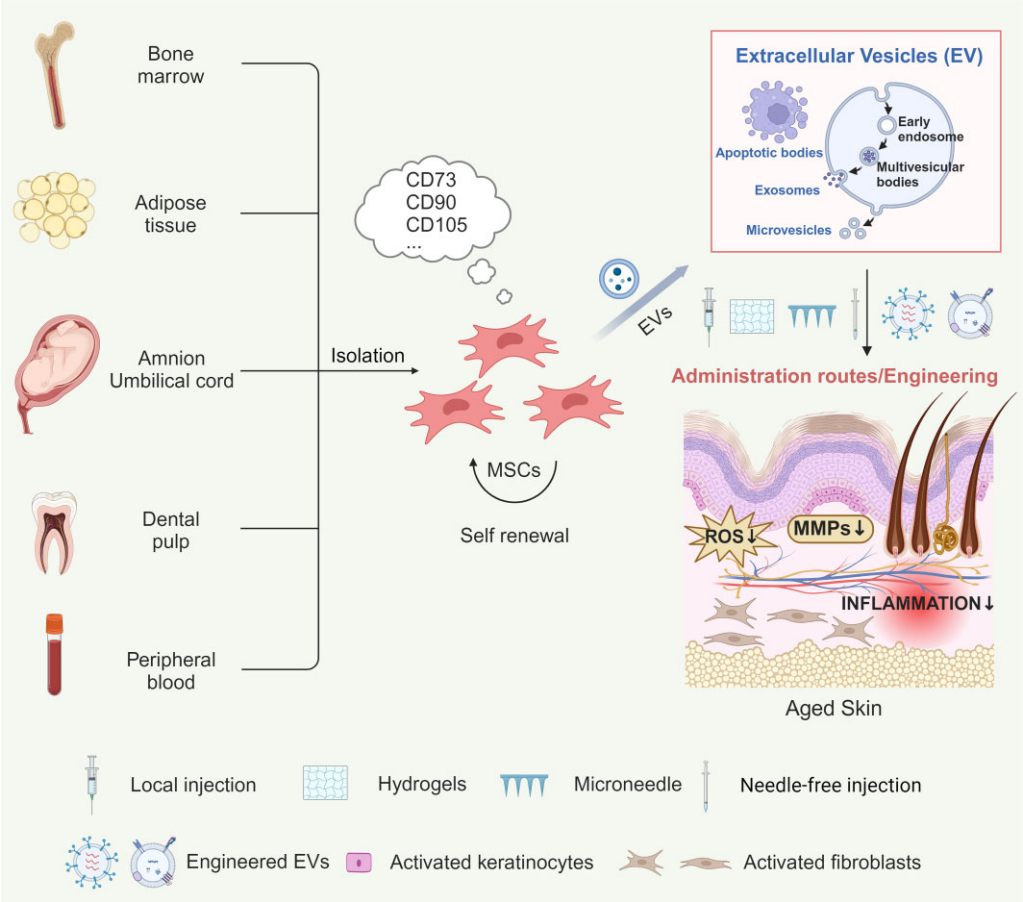


Figure 3. Mechanisms and therapeutic methods of MSC-EVs for delaying skin aging. MSCs are a type of multipotent stem cell with self-renewal capabilities and can be isolated from bone marrow, adipose tissue, amnion umbilical cord, dental pulp, and peripheral blood. CD73, CD90, CD105, etc. are surface markers of MSCs. MSC-EVs delay skin aging by activating keratinocytes and fibroblasts, promoting their proliferation and migration, removing MMPs, reducing the generation of ROS, and regulating the inflammatory response. The administration routes of MSC-EVs include local injection, hydrogels, microneedle, and needle-free injection. In addition, engineered EVs can carry specific cargoes for targeted drug delivery.

undergoes a solution-to-gel transition when the pH changes from high to neutral, resulting in immediate gelation when introduced into physiological systems. Following the topical *in vivo* administration of fluorescently labeled EVs incorporated into UPy-hydrogel, the presence of EVs was still observed within the UPy-hydrogel after a period of 3 days. In the absence of the hydrogel, EV was internalized by adipose and skin tissues near the injection site.

Chitosan hydrogel (CS), with its thermosensitive, loose and porous structural properties, has been used as a carrier for the slow release of drugs in several studies [155, 156]. A study by Zhao *et al.* [157] suggests that CS hydrogel-encapsulated EVs can improve skin aging by enhancing the function of aged dermal fibroblasts. According to their findings, CS hydrogel-incorporated EVs (CS-EVs) target the dermal fibroblasts with replicative senescence, promote cell proliferation, and enhance ECM protein synthesis in aged cells and the upregulation of MMPs *in vitro*. Following the subcutaneous injection of CS-EVs, the aging skin tissues exhibited a state of rejuvenation, characterized by a notable increase in collagen expression, a decrease in the expression of SASP-related factors, and the restoration of tissue structures.

Lipodystrophy may manifest as a reduction in subcutaneous fat, particularly in areas such as the face, arms, and legs. This can lead to thinner and looser-looking skin, potentially affecting the overall appearance. Injections of hydrogel for volume filling can go some way to making a person look younger. HA hydrogels (HA-Gels) are often used as soft tissue fillers in the face, lips, and buttocks to increase their size and make them more aesthetically pleasing. According to You *et al.* [158], stem cell-derived EV-containing HA gels (EV-HA-Gels) could be used as dermal fillers. EV-HA-Gels induced the overexpression of CD301b on macrophages. The authors provided evidence of the ability of EV-HA-Gels to induce the upregulation of CD301b expression on macrophages. Certain miRNAs, specifically let-7b-5p and miR-243p, were found to play a role in the impact of EV-HA-Gels on enhancing fibroblast proliferation in the dermis region. *In vivo* experiments demonstrated a significant upregulation of collagen synthesis in the treated dermis when compared to dermis treated with HA-Gel alone, with a 2.4-fold increase. Furthermore, these elevated collagen levels were sustained for a minimum of 24 weeks in the dermis.

Microneedles (MN)

MN are miniature-sized structures capable of delivering drugs to deeper layers of the skin without compromising the skin barrier. Micro-scale size endows them with many advantages over hypodermic needles, including painlessness, minimal invasiveness, and convenient operation [159]. In addition, previous reports have shown that MN treatment alone aids collagen neovascularization. Hong *et al.* [160] reported that wrinkles and skin roughness

Table 1. Functions and therapeutic mechanisms of MSC-EVs against skin aging.

Source	Nomenclature	Model	Function	Therapeutic mechanism	References
Human ADSCs	Exosomes	HaCaTs	Improving wound healing	Highly-expressed lncRNA FOXD2-AS1 in ADSC-Exos promote HaCaTs migration and proliferation via miR-185–5p/ROCK2 axis	[115]
Human uMSCs	Exosomes	H_2O_2-HaCaTs and mouse	Improving wound healing	Suppress the nuclear translocation of AIF and the excessive activation of PARP-1, up-regulate CD31, cytokeratin 10 and down-regulate alpha-SMA	[116]
Human ADSCs	Exosomes	HDFs and mouse	Improving wound healing	Promote HDFs proliferation and migration and synthesize collagen I/III via the PI3K/Akt signaling pathway	[117]
Human BMSCs	Exosomes	HaCaTs,HDFs, and mouse	Improving wound healing	Promote HaCaTs and HDFs proliferation, down-regulate the expression of TGF-beta1, Smad2, Smad3, and Smad4, and up-regulate the expression of TGF-beta3 and Smad7	[118]
Human ADSCs	EVs	UVB-induced HDFs	Against photoaging	Increase TIMP-1, TGF-beta1, and collagen types I, II, III, and V, and elastin, reduce MMP-1, -2, -3, and -9	[23]
Human uMSCs	EVs	UVB-induced HDFs	Against photoaging	Reduce MMP-1 and the percentage of senescent cells, increase collagen I	[123]
Human ADSCs	Exosomes	UVB-induced HDFs and mouse	Against photoaging	Reduce MMP-1 and increase type I procollagen via MAPK/AP-1 signaling pathway and TGF-beta/Smad pathway	[124]
Human uMSCs	Exosomes	H_2O_2-Keratinocytes and mouse	Against oxidative stress	Up-regulate FRAP, GPx and SOD concentrations to reduce ROS production via NRF2 signaling pathway	[130]
Human ADSCs	Exosomes	HaCaTs	Against oxidative stress	Reduce ROS and increase collagen remodeling	[131]
Human ADSCs	Exosomes	EPCs	Against oxidative stress	Overexpression of Nrf2 in ADSC-Exos reduces ROS level	[132]
Human uMSCs	Exosomes	UV-induced HaCaTs	Against oxidative stress	HuMSC-Exos-delivered 14–3-3ζ protein promotes SIRT1 expression to reduce ROS	[133]
Human ESCs	Exosomes	Psoriasis mouse	Anti-inflammatory	Reduce IL-17, IL-23, and C5b-9	[142]
Human ADSCs	Exosomes	Atopic dermatitis mouse	Anti-inflammatory	Reduce $CD86^+$, $CD206^+$, eosinophils, and mast cells, inhibit TNF-alpha, IL-23, IL-31, and IL-4 expression	[143]
Human ADSCs	Exosomes	HDFs	Anti-inflammatory	lncRNA GAS5 in ADSC-Exos reduces IFNalpha, IL1beta, and TNFα levels by suppressing toll-like receptor 7 expression	[144]
Human BMSCs	Exosomes	Macrophages and mouse	Anti-inflammatory	Transform M1 macrophages to M2 macrophages through PI3K/AKT signaling pathway, promote angiogenesis and IL-10, suppress IL-1beta and TNF-α	[145]
Selenium-pretreated human BMSCs	Exosomes	HDFs, macrophages and mouse	Anti-inflammatory	Reduce p16, p21, and ROS, inhibit TNF-α, IL-6, and IL-8, promote TGF-beta1, IL-10, and M1 to M2 polarization of macrophages, promote angiogenesis, migration, and wound closure	[146]

ESCs, Embryonic stem cells; EPCs, endothelial progenitor cells.

values were reduced in photoaged mice after MN treatment, and histological examination showed a slight increase in collagen and elastin fibers.

Cao *et al.* [161] investigated the effect of MN combined with ADSCs-EVs on skin aging. They found reduced epidermal thickness and enhanced skin barrier function at sites treated with MN alone or MN + EVs compared to the untreated sites. However, the MN + EVs group showed the fewest wrinkles, the highest collagen density, and the most organized collagen fibers among the three groups. Three days after treatment, $CD11b^+$ cell infiltration was lower in the MN + EVs group than in the MN group. These results suggest that MN treatment alone can improve epidermal structure and function in photoaged skin. In addition, the combination with ADSCs-EVs accelerates the recovery of inflammation caused by MN and increases collagen content. You *et al.* [162] designed a MN patch system loaded with COL1A1-EV and HA by a micro-molding method, and named this system COL1A1-EV MN. For delivery into tissue, COL1A1-EV MN patches were pressed into the dorsal skin of mice, and the MN base was removed after 15 min. During this period, the MN dissolved completely, with no visible skin irritation or marking at the site of administration. They found that EVs administered using MN patches were significantly more homogeneous and durable compared to insulin syringes.

The marine sponge Haliclona sp. spicules (SHS), composed of silicious oxeas, has the ability to penetrate the skin through simple massage, resulting in the creation of over 1000 microchannels (with a depth of 42.2 ± 14.9 μm) per mm^2. According to recent reports, this distinctive characteristic of SHS renders it a potentially advantageous MN system for augmenting the skin's absorption of hydrophilic macromolecules. Significantly, SHS exhibits the capability to be employed at any desired location on the skin, unlike previous MN patches that were restricted to limited and flat

areas [163]. Considering the many advantages of SHS, Zhang *et al.* [164] explored whether the use of SHS could enhance the cutaneous delivery of HuMSC-Exos. *In vitro* experiments revealed that HuMSC-Exos exhibited limited ability to permeate pig skin. Nevertheless, the utilization of SHS resulted in a 5.87-fold enhancement in exosome uptake through the creation of microchannels. Subsequent *in vivo* experiments conducted on photoaged mice demonstrated that the concurrent administration of HuMSC-Exos and SHS yielded noteworthy anti-photoaging outcomes, encompassing the reduction of micro-wrinkles, mitigation of histopathological alterations, and stimulation of ECM component expression. Conversely, the sole application of HuMSC-Exos exhibited considerably weaker effects. The skin irritation test demonstrated that the co-administration of HuMSC-Exos and SHS resulted in a minor degree of irritation, which promptly resolved.

Targeting strategy and EVs

Targeted strategies for drug delivery are a wonderful aspiration for precision medicine, which not only avoids drug waste but also drastically reduces drug side effects. Previous studies have shown that EVs have a strong targeting ability, mainly due to plasma membrane targeting, higher-order oligomerization, and protein modification (e.g. myristoylation, prenylation, and palmitoylation) [165–167].

Liposomes, tiny lipid vesicles surrounded by a membrane bilayer composed primarily of phospholipids and cholesterol, can serve as an important drug delivery system because of their amphiphilic nature, but their non-targeting, low potency, and short half-life hinder their translational applications [168, 169]. These deficiencies can be significantly mitigated by appropriate functionalization of these drug carriers with biological entities possessing the targeting characteristics and biological constructs of EVs. For example, the use of specific cell membrane-encapsulated nanovesicles may offer promising delivery strategies for clinical applications. Hu *et al.* [170] have devised a top-down methodology for the concealment of poly (lactic-co-glycolic acid) (PLGA) NPs using natural erythrocyte membranes. The resultant NPs, camouflaged with membranes, have demonstrated remarkable blood residence capabilities. The incorporation of membrane lipids and associated membrane proteins has effectively hindered macrophage-mediated particle clearance. Moreover, the researchers employed a technique to conceal PLGA NPs using plasma membranes derived from human platelets, thereby conferring upon these NPs the immunomodulatory and adhesive antigens typically found on platelets. Subsequently, these NPs were utilized for the purpose of restoring impaired blood vessels and addressing systemic infections induced by opportunistic pathogens within a living organism [171]. Boada *et al.* [172] effectively integrated proteins obtained from leukocyte plasma membrane into lipid NPs, thereby creating biomimetic vesicles with the purpose of specifically targeting inflamed vasculature. Evers *et al.* [173] introduced novel types of hybrid NPs that combine EVs and liposomes. The authors provided evidence in their study to support the notion that these hybrid NPs, which incorporate target genes and EV-surface markers, facilitate the precise delivery of genes to particular cells. Zhou *et al.* [174] isolated tumor-derived extracellular vesicles from hepatocellular carcinoma cells and replaced their contents to achieve targeting of tumor cells.

In addition to utilizing the characteristics of EVs to enhance liposome targeting, studies have also been conducted to facilitate EV targeting to specific receptor cells through exogenous peptides, proteins, or lipid-modified designed EVs [175]. Among the specific preparation strategies, there are broadly two types of schemes: genetic modification of parental cells and chemical modification of preformed EVs [176]. The former can be accomplished by cloning protein sequences and connecting protein sequences with targeting potential to specific protein sequences, so as to "anchor" specific proteins to the surface of vesicle membranes. As an illustration, the C1C2 domain is situated at the C-terminus of the fusion sequence. Upon expression in host cells, the signal facilitates the transportation of the complete protein into the secreted EV, thereby orienting the N-terminal region towards the outer surface of the EV. This method has been used to develop tumor vaccines and enhance the targeting of drug delivery systems [177–179]. Given the ability of rabies viral glycoprotein to specifically bind acetylcholine receptor [3], Alvarez-Erviti *et al.* [180] engineered dendritic cells to express exosome membrane protein Lamp2b (a protein found abundantly in exosomal membranes) fused with neuron-specific rabies viral glycoprotein peptide, thereby achieving the targeting of vesicles to the nervous system. As for the second method, Antes *et al.* [181] describe a protocol to increase EV targeting, in their study, by means of an EV membrane-anchoring platform called "cloaking" (consisting of three components, including a DMPE phospholipid membrane anchor, a polyethylene glycol spacer and a conjugated streptavidin platform molecule, to which any biotinylated molecule can be coupled for EV decoration) to enhance vesicle uptake in cells of interest by embedding tissue-specific antibodies or homing peptides directly *in vitro* on the surface of the EV membrane. Cao *et al.* [182] used RGD-modified EV as a carrier to achieve the targeting of photothermal effectors to tumor cells. Qi *et al.* [183] developed a bifunctional superparamagnetic NP cluster based on exosomes as a targeted drug delivery carrier for cancer treatment. This exosome-based drug delivery carrier is superparamagnetic at room temperature and responds more strongly to an external magnetic field than a single superparamagnetic NP. These characteristics enable exosomes to be isolated from the blood and target diseased cells.

To date, no additional EV-targeting enhancements have been reported for the treatment of skin aging. We believe that this may be a future direction for research to enhance drug efficacy and avoid side effects, e.g. targeting specific types of reparative cells and providing a program that is specific to these types of cells.

Enrichment of specific inclusions in EV cargoes

Due to the increasing research on the mechanism of aging, it is expected that the characteristic of targeted transport of EVs will be utilized to transport specific fragments of nucleic acids or proteins into the target cytoplasm [184, 185]. Enrichment of EV inclusions is also an important modification option for EVs.

Zhang *et al.* [125] found that circ_0011129, a non-coding circular RNA, functions as a miR-6732-5p adsorption sponge, thereby impeding the degradation of type I collagen and the aggregation of elastin in a UVA-induced photoaging model of human dermal fibroblast cells. To improve the *in vivo* stability and delivery efficiency of circRNAs, the authors prepared extracellular vesicles loaded with circ_0011129 (3D-circc-sEV) by overexpressing circ_0011129 in ADSCs and culturing them in a 3D bioreactor. Finally, they demonstrated that 3D-circ-sEVs possess the ability to impede the cellular photoaging process and safeguard cells against UVA radiation-induced harm as well as in a model of oxidative stress induced by H_2O_2. Gao *et al.* [124] utilized lentivirus infection to obtain ADSCs and exosomes (OE-EX) that exhibited high expression of miR-1246. They subsequently investigated

the potential anti-photoaging effect of OE-EX on human skin fibroblasts (HSFs) and Kunming mice. The findings demonstrated that OE-EX displayed a more pronounced anti-photoaging effect, which was attributed to its ability to significantly reduce MMP-1 levels through the inhibition of the MAPK/AP-1 signaling pathway. Additionally, OE-EX effectively enhanced the secretion of type I collagen by activating the TGF-beta/Smad pathway, thereby preventing UV-induced IκB-α degradation and NF-κB overexpression, ultimately exerting an anti-inflammatory effect.

Due to size constraints, studies of EVs cargoes usually focus on nucleic acids with small fragments (10–20 nt range), such as miRNAs and small interfering RNAs. Larger nucleic acids such as mRNA are more difficult to load into EVs [186]. Yang *et al.* [187] conducted a study wherein they transfected different source cells with plasmid DNAs and subsequently stimulated the cells using a localized and temporary electrical stimulus. This stimulus facilitated the release of exosomes containing transcribed mRNAs and targeting peptides. In comparison to bulk electroporation and alternative methods for exosome production, cellular nanoporation resulted in a significantly higher yield of exosomes, up to a 50-fold increase, and a substantial increase of >103-fold in exosomal mRNA transcripts. This effect was observed even in cells with initially low levels of exosome secretion. Using this approach, called cellular nanoporation, You *et al.* [162] enriched mRNA encoding extracellular-matrix COL1A1 into EVs, which induced the formation of collagen grafts and reduced the formation of wrinkles in collagen-depleted dermal tissues of mice with photoaged skin.

Other administration routes

The needle-free injector has gained significant popularity in the administration of local anesthesia and vaccines in humans, primarily owing to its high levels of safety and effectiveness, particularly among individuals with a fear of needles [188]. The efficacy of needleless syringes in delivering exosomal treatments was assessed by Hu *et al.* [188]. This method involves the pneumatic acceleration of exosomal solutions into the dermis of the skin, resulting in reduced damage and pain, enhanced penetration and absorption, and improved suitability for cosmetic applications compared to conventional syringes. To investigate the impact of these injection methods on the distribution of extracellular vesicles in the dermis, the researchers employed a DID labeling technique. The findings indicate that the administration of exosomes via syringe injection induces the aggregation of concentrated substances and inflammatory cells. Conversely, needleless syringes do not exhibit these limitations. Furthermore, the minor dermal injuries caused by needleless syringes are conducive to the synthesis of collagen in the skin (Fig. 3) [189].

Conclusion and prospects

Skin aging can be divided into intrinsic aging and extrinsic aging, which involves oxidative stress, telomere shortening, mitochondrial, melatonin, and miRNA changes, inflammaging, immunosenescence, and autophagy. There are several positive aspects of MSC-EVs in delaying skin aging. Firstly, MSC-EVs accelerate the proliferation and migration of HDFs and keratinocytes and inhibit apoptosis, thus promoting skin tissue regeneration. Secondly, UV radiation activates MAPK and NF-kB signaling pathways, leading to an increase in MMPs. MSC-EVs inhibit the activation of these signaling pathways to reverse the level of MMPs, thus promoting ECM synthesis. Furthermore, MSC-EVs protects cells from oxidative damage caused by excessive oxidative stress through activating the antioxidant defense system. In addition, MSC-EVs possess anti-inflammatory and immunomodulatory properties and inhibit the release of inflammatory mediators. As for therapeutic methods, local injection is the most common administration method of MSC-EVs, and hydrogel preparation, MN, and needle-free injection combination therapy has its own advantages. Engineered EVs can carry specific cargoes for drug delivery according to their targeting capabilities.

Compared to traditional stem cell therapy, MSC-EVs offer several distinct advantages. Firstly, conventional stem cell therapy often entails the direct transplantation of exogenous cells into the patient's body, a procedure that carries the risk of immune rejection reactions. In contrast, EVs exhibit a lower propensity for immune reactions due to their diminished immunogenicity, which renders them more readily accepted by the patient's immune system, thereby reducing the likelihood of rejection [190, 191]. Secondly, the preparation of EVs is relatively straightforward, achievable through *in vitro* culture, and conducive to cryopreservation and long-term storage. These attributes greatly facilitate their clinical application and enhance treatment feasibility [192, 193]. Additionally, EVs convey a diverse array of bioactive molecules encompassing proteins, nucleic acids, lipids, and cytokines. These molecular constituents play pivotal roles in modulating immune responses, facilitating tissue repair, and exerting anti-inflammatory effects, among other functions. Consequently, EVs hold substantial promise in the treatment of diverse diseases [185, 194]. Moreover, EV-based therapy affords expanded prospects for personalized medicine, allowing for customization according to individual patient conditions and needs, thereby achieving more precise therapeutic outcomes. In summary, MSC-EVs represent promising candidate drugs for addressing skin aging and related conditions.

Nonetheless, despite the remarkable strides made in this field, numerous pivotal issues remain that require attention. Firstly, the composition and effectiveness of EV therapy may be influenced by various factors, including donor variability, culture conditions, isolation and purification methods, and more. This variability introduces uncertainty to the therapeutic outcomes and poses a significant challenge to standardizing the quality of EVs. The preparation, storage, and application of EV therapy requires specialized laboratory conditions and technology. Large-scale production can significantly increase costs, especially in healthcare systems with limited resources. Proper storage conditions are crucial for maintaining the activity and quality of EVs, often necessitating the use of specialized freezing equipment and storage facilities. During the application phase, introducing EV therapy to patients requires adherence to high standards of laboratory procedures and technology to ensure the safety and effectiveness of the treatment [193, 195].

Secondly, determining the optimal dosage and most suitable administration methods for EV therapy remains a complex task. Insufficient dosage may fail to produce therapeutic effects, while excessive dosage may pose unnecessary risks. Further research is needed to clarify the optimal dosage of EVs and the most effective methods for their delivery into the patient's body. Regarding human skin, the delivery of EVs may face some obstacles. The barrier function of the skin's surface stratum corneum may limit the penetration of EVs. The immune system present in the skin may lead to immune rejection reactions against EVs [189, 196]. Factors such as hair follicles, skin pH, humidity, and other physicochemical properties can also affect the penetration efficiency of EVs.

Moreover, although MSC-EVs are generally considered relatively safe, safety considerations are crucial in clinical applications [89]. This becomes particularly prominent in the context of long-term treatments, requiring enhanced review and monitoring of potential adverse reactions. During the clinical application process, careful monitoring of patients is necessary to ensure that the treatment does not trigger unexpected immune responses or other adverse events. Meanwhile, more in-depth research is needed regarding the sustainability and long-term stability of the therapeutic effects of EVs. For instance, it is crucial to investigate whether EVs, after being delivered into the skin, can remain stably present and exert continuous effects. The clearance and metabolism mechanisms of EVs by the skin may impact the sustainability of the treatment. A recent study indicates that encapsulating COL1A1-mRNA into EVs can reduce the formation of wrinkles in photoaged mice. After low-dose injection, COL1A1 was significantly increased in local skin tissue at 12 h, peaked on the fourth day, and returned to baseline levels by day 30. However, EVs delivered via HA MNs exhibited better dispersion in the dermal layer, leading to a substantial reduction in wrinkles for up to 70 days with the initial dosage [162].

While the therapeutic potential of EVs in combating skin aging is substantial, further in-depth studies are imperative to elucidate the underlying mechanisms comprehensively. Formal clinical adoption of MSC-EVs for the treatment of skin aging requires continued research efforts.

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Author contributions

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Conflict of interest

The authors declare no competing interests.

References

1. Byrd AL, Belkaid Y, Segre JA. The human skin microbiome. *Nat Rev Micro* 2018;**16**:143–55. https://doi.org/10.1038/nrmicro.2017.157.
2. Boroni M, Zonari A, Reis De Oliveira C *et al.* Highly accurate skin-specific methylome analysis algorithm as a platform to screen and validate therapeutics for healthy aging. *Clinical Epigenetics* 2020;**12**:105. https://doi.org/10.1186/s13148-020-00899-1.
3. Freitas R, Martins A, Silva J *et al.* Highlighting the biological potential of the brown seaweed fucus spiralis for skin applications. *Antioxidants (Basel, Switzerland)* 2020;**9**:611. https://doi.org/10.3390/antiox9070611.
4. Lee D, Kim Y, Jo H *et al.* The anti-inflammatory effect of Aptamin C on House dust mite extract-induced inflammation in keratinocytes via regulation of IL-22 and GDNF production. *Antioxidants (Basel, Switzerland)* 2021;**10**:945. https://doi.org/10.3390/antiox10060945.
5. Cho HS, Lee MH, Lee JW *et al.* Anti-wrinkling effects of the mixture of vitamin C, vitamin E, pycnogenol and evening primrose oil, and molecular mechanisms on hairless mouse skin caused by chronic ultraviolet B irradiation. *Photodermatol Photoimmunol Photomed* 2007;**23**:155–62. https://doi.org/10.1111/j.1600-0781.2007.00298.x.
6. Knott A, Achterberg V, Smuda C *et al.* Topical treatment with coenzyme Q10-containing formulas improves skin's Q10 level and provides antioxidative effects. *Biofactors* 2015;**41**:383–90. https://doi.org/10.1002/biof.1239.
7. Cao C, Xiao Z, Tong H *et al.* Oral intake of chicken bone collagen peptides anti-skin aging in mice by regulating collagen degradation and synthesis, inhibiting inflammation and activating lysosomes. *Nutrients* 2022;**14**:1622. https://doi.org/10.3390/nu14081622.
8. Papakonstantinou E, Roth M, Karakiulakis G. Hyaluronic acid: A key molecule in skin aging. *Dermato-endocrinology* 2012;**4**:253–8. https://doi.org/10.4161/derm.21923.
9. Polo-Castellano C, Mateos RM, Visiedo F *et al.* Optimizing an enzymatic extraction method for the flavonoids in moringa (Moringa oleifera Lam.) leaves based on experimental designs methodologies. *Antioxidants (Basel, Switzerland)* 2023;**12**:369. https://doi.org/10.3390/antiox12020369.
10. Keshtkar S, Azarpira N, Ghahremani MH. Mesenchymal stem cell-derived extracellular vesicles: novel frontiers in regenerative medicine. *Stem Cell Res Ther* 2018;**9**:63. https://doi.org/10.1186/s13287-018-0791-7.
11. Deng L, Ren R, Liu Z *et al.* Stabilizing heterochromatin by DGCR8 alleviates senescence and osteoarthritis. *Nat Commun* 2019;**10**:3329. https://doi.org/10.1038/s41467-019-10831-8.
12. Kim H, Lee MJ, Bae E-H *et al.* Comprehensive molecular profiles of functionally effective MSC-derived extracellular vesicles in immunomodulation. *Mol Ther* 2020;**28**:1628–44. https://doi.org/10.1016/j.ymthe.2020.04.020.
13. Dyball LE, Smales CM. Exosomes: biogenesis, targeting, characterization and their potential as "Plug & Play" vaccine platforms. *Biotechnol J* 2022;**17**: e2100646. https://doi.org/10.1002/biot.202100646.
14. Fu M, Gu J, Jiang P *et al.* Exosomes in gastric cancer: roles, mechanisms, and applications. *Mol Cancer* 2019;**18**:41. https://doi.org/10.1186/s12943-019-1001-7.
15. Xie S, Zhang Q, Jiang L. Current knowledge on exosome biogenesis, cargo-sorting mechanism and therapeutic implications. *Membranes* 2022;**12**:498. https://doi.org/10.3390/membranes12050498.
16. Schünke H, Göbel U, Dikic I *et al.* OTULIN inhibits RIPK1-mediated keratinocyte necroptosis to prevent skin inflammation in mice. *Nat Commun* 2021;**12**:5912. https://doi.org/10.1038/s41467-021-25945-1.
17. Arda O, Göksügür N, Tüzün Y. Basic histological structure and functions of facial skin. *Clin Dermatol* 2014;**32**:3–13. https://doi.org/10.1016/j.clindermatol.2013.05.021.

18. Bollag WB, Aitkens L, White J *et al.* Aquaporin-3 in the epidermis: more than skin deep. *Am J Physiol Cell Physiol* 2020;**318**: C1144–C1153. https://doi.org/10.1152/ajpcell.00075.2020.
19. Denis J-F, Lévesque M, Tran SD *et al.* Axolotl as a model to study scarless wound healing in vertebrates: role of the transforming growth factor beta signaling pathway. *Adv Wound Care* 2013;**2**:250–60. https://doi.org/10.1089/wound.2012.0371.
20. Fu H, Zhang Y, An Q *et al.* Anti-photoaging effect of rhodiola rosea fermented by Lactobacillus plantarum on UVA-damaged fibroblasts. *Nutrients* 2022;**14**:2324. https://doi.org/10.3390/nu14112324.
21. Younis S, Deeba F, Fatima Saeed R *et al.* Regulation of cell cycle and differentiation markers by pathogenic, non-pathogenic and opportunistic skin bacteria. *Saudi J Biolog Sci* 2022;**29**:1717–29. https://doi.org/10.1016/j.sjbs.2021.10.058.
22. Gu Y, Han J, Jiang C *et al.* Biomarkers, oxidative stress and autophagy in skin aging. *Ageing Res Rev* 2020;**59**:101036. https://doi.org/10.1016/j.arr.2020.101036.
23. Choi JS, Lee Cho W, Choi YJ *et al.* Functional recovery in photo-damaged human dermal fibroblasts by human adipose-derived stem cell extracellular vesicles. *J Extracell Vesicles* 2019;**8**:1565885. https://doi.org/10.1080/20013078.2019.1565885.
24. Bielach-Bazyluk A, Zbroch E, Mysliwiec H *et al.* Sirtuin 1 and skin: implications in intrinsic and extrinsic aging-A systematic review. *Cells* 2021;**10**:813. https://doi.org/10.3390/cells10040813.
25. Low E, Alimohammadiha G, Smith LA *et al.* How good is the evidence that cellular senescence causes skin ageing? *Ageing Res Rev* 2021;**71**:101456. https://doi.org/10.1016/j.arr.2021.101456.
26. Iwasaki O, Tanizawa H, Kim K-D *et al.* Involvement of condensin in cellular senescence through gene regulation and compartmental reorganization. *Nat Commun* 2019;**10**:5688. https://doi.org/10.1038/s41467-019-13604-5.
27. Chen L, Liao F, Wu J *et al.* Acceleration of ageing via disturbing mTOR-regulated proteostasis by a new ageing-associated gene PC4. *Aging Cell* 2021;**20**: e13370. https://doi.org/10.1111/acel.13370.
28. Blair MJ, Jones JD, Woessner AE *et al.* Skin structure-function relationships and the wound healing response to intrinsic aging. *Adv Wound Care* 2020;**9**:127–43. https://doi.org/10.1089/wound.2019.1021.
29. Chambers ES, Vukmanovic-Stejic M.. Skin barrier immunity and ageing. *Immunology* 2020;**160**:116–25. https://doi.org/10.1111/imm.13152.
30. Liu T, Li N, Yan Y-Q *et al.* Recent advances in the anti-aging effects of phytoestrogens on collagen, water content, and oxidative stress. *Phytother Res* 2020;**34**:435–47. https://doi.org/10.1002/ptr.6538.
31. Monteleone P, Mascagni G, Giannini A *et al.* Symptoms of menopause—global prevalence, physiology and implications. *Nat Rev Endocrinol* 2018;**14**:199–215. https://doi.org/10.1038/nrendo.2017.180.
32. Krutmann J, Schikowski T, Morita A *et al.* Environmentally-induced (Extrinsic) skin aging: exposomal factors and underlying mechanisms. *J Invest Dermatol* 2021;**141**:1096–103. https://doi.org/10.1016/j.jid.2020.12.011.
33. Wang F, Smith NR, Tran BAP *et al.* Dermal damage promoted by repeated low-level UV-A1 exposure despite tanning response in human skin. *JAMA Dermatology* 2014;**150**:401–6. https://doi.org/10.1001/jamadermatol.2013.8417.
34. Bernerd F, Passeron T, Castiel I *et al.* The damaging effects of long UVA (UVA1) rays: A major challenge to preserve skin health and integrity. *Int J Mol Sci* 2022;**23**:8243. https://doi.org/10.3390/ijms23158243.
35. Pelclova D, Navratil T, Kacerova T *et al.* NanoTiO(2) sunscreen does not prevent systemic oxidative stress caused by UV radiation and a Minor amount of NanoTiO(2) is absorbed in humans. *Nanomaterials (Basel)* 2019;**9**:888. https://doi.org/10.3390/nano9060888.
36. Kim DJ, Iwasaki A, Chien AL *et al.* UVB-mediated DNA damage induces matrix metalloproteinases to promote photoaging in an AhR- and SP1-dependent manner. *JCI Insight* 2022;**7**:e156344. https://doi.org/10.1172/jci.insight.156344.
37. Karapetsas A, Voulgaridou G-P, Iliadi D *et al.* Honey extracts exhibit cytoprotective properties against UVB-induced photodamage in Human experimental skin models. *Antioxidants (Basel, Switzerland)* 2020;**9**:566. https://doi.org/10.3390/antiox9070566.
38. Georgescu SR, Mitran CI, Mitran MI *et al.* Oxidative stress in cutaneous lichen planus-A narrative review. *J Clin Med* 2021;**10**:2692. https://doi.org/10.3390/jcm10122692.
39. Tsuchida K, Kobayashi M. Ultraviolet A irradiation induces ultraweak photon emission with characteristic spectral patterns from biomolecules present in human skin. *Sci Rep* 2020;**10**:21667. https://doi.org/10.1038/s41598-020-78884-0.
40. Lan Y, Wang Y, Lu H. Opsin 3 is a key regulator of ultraviolet A-induced photoageing in human dermal fibroblast cells. *Br J Dermatol* 2020;**182**:1228–44. https://doi.org/10.1111/bjd.18410.
41. Alam MB, Chowdhury NS, Sohrab MH *et al.* Cerevisterol alleviates inflammation via suppression of MAPK/NF-κB/AP-1 and activation of the Nrf2/HO-1 signaling cascade. *Biomolecules* 2020;**10**:199. https://doi.org/10.3390/biom10020199.
42. Pittayapruek P, Meephansan J, Prapapan O *et al.* Role of matrix metalloproteinases in photoaging and photocarcinogenesis. *Int J Mol Sci* 2016;**17**:868. https://doi.org/10.3390/ijms17060868.
43. Han S, Ballinger E, Choung S-Y *et al.* Anti-photoaging effect of hydrolysates from Pacific whiting skin via MAPK/AP-1, NF-kappaB, TGF-beta/smad, and nrf-2/HO-1 signaling pathway in UVB-induced Human dermal fibroblasts. *Mar Drugs* 2022;**20**:308. https://doi.org/10.3390/md20050308.
44. Xu D, Li C, Zhao M. Attenuation of UV-induced skin photoaging in rats by walnut protein hydrolysates is linked to the modulation of MAPK/AP-1 and TGF-beta/smad signaling pathways. *Food Funct* 2022;**13**:609–23. https://doi.org/10.1039/d1fo02598h.
45. Aguilera P, Whalen J, Minguet C *et al.* The nuclear pore complex prevents sister chromatid recombination during replicative senescence. *Nat Commun* 2020;**11**:160. https://doi.org/10.1038/s41467-019-13979-5.
46. Xu X, Li Y, Bharath SR *et al.* Structural basis for reactivating the mutant TERT promoter by cooperative binding of p52 and ETS1. *Nat Commun* 2018;**9**:3183. https://doi.org/10.1038/s41467-018-05644-0.
47. Victorelli S, Lagnado A, Halim J *et al.* Senescent human melanocytes drive skin ageing via paracrine telomere dysfunction. *EMBO J* 2019;**38**: e101982. https://doi.org/10.15252/embj.2019101982.
48. Jia Y, Mao Q, Yang J *et al.* (-)-Epigallocatechin-3-gallate protects Human skin fibroblasts from ultraviolet a induced photoaging. *Clin Cosmet Investig Dermatol* 2023;**16**:149–59. https://doi.org/10.2147/CCID.S398547.
49. Marion RM, Strati K, Li H *et al.* Telomeres acquire embryonic stem cell characteristics in induced pluripotent stem cells. *Cell Stem Cell* 2009;**4**:141–54. https://doi.org/10.1016/j.stem.2008.12.010.

50. Flores I, Cayuela ML, Blasco MA. Effects of telomerase and telomere length on epidermal stem cell behavior. *Science* 2005;**309**:1253–6. https://doi.org/10.1126/science.1115025.
51. Ball JM, Chen S, Li W. Mitochondria in cone photoreceptors act as microlenses to enhance photon delivery and confer directional sensitivity to light. *Sci Adv* 2022;**8**:eabn2070. https://doi.org/10.1126/sciadv.abn2070.
52. Watanabe M, Natsuga K, Nishie W *et al.* Type XVII collagen coordinates proliferation in the interfollicular epidermis. *eLife* 2017;**6**: e26635. https://doi.org/10.7554/eLife.26635.
53. Sreedhar A, Aguilera-Aguirre L, Singh KK. Mitochondria in skin health, aging, and disease. *Cell Death Dis* 2020;**11**:444. https://doi.org/10.1038/s41419-020-2649-z.
54. Xu Y, Qi F, Mao H *et al.* In-situ transfer vat photopolymerization for transparent microfluidic device fabrication. *Nat Commun* 2022;**13**:918. https://doi.org/10.1038/s41467-022-28579-z.
55. Chen X, Su J, Wang R *et al.* Structural optimization of cannabidiol as multifunctional cosmetic raw materials. *Antioxidants (Basel, Switzerland)* 2023;**12**:314. https://doi.org/10.3390/antiox12020314.
56. Berneburg M, Plettenberg H, Medve-König K *et al.* Induction of the photoaging-associated mitochondrial common deletion in vivo in normal human skin. *J Invest Dermatol* 2004;**122**:1277–83. https://doi.org/10.1111/j.0022-202X.2004.22502.x.
57. Schroeder P, Gremmel T, Berneburg M *et al.* Partial depletion of mitochondrial DNA from human skin fibroblasts induces a gene expression profile reminiscent of photoaged skin. *J Invest Dermatol* 2008;**128**:2297–303. https://doi.org/10.1038/jid.2008.57.
58. Arslanbaeva L, Tosi G, Ravazzolo M *et al.* UBIAD1 and CoQ10 protect melanoma cells from lipid peroxidation-mediated cell death. *Redox Biol* 2022;**51**:102272. https://doi.org/10.1016/j.redox.2022.102272.
59. López-Lluch G. Coenzyme Q homeostasis in aging: response to non-genetic interventions. *Free Radical Biol Med* 2021;**164**:285–302. https://doi.org/10.1016/j.freeradbiomed.2021.01.024.
60. Marcheggiani F, Cirilli I, Orlando P *et al.* Modulation of Coenzyme Q(10) content and oxidative status in human dermal fibroblasts using HMG-CoA reductase inhibitor over a broad range of concentrations. From mitohormesis to mitochondrial dysfunction and accelerated aging. *Aging* 2019;**11**:2565–82. https://doi.org/10.18632/aging.101926.
61. Bešlić I, Lugović-Mihić L, Vrtarić A *et al.* Melatonin in dermatologic allergic diseases and other skin conditions: current trends and reports. *Int J Mol Sci* 2023;**24**:4039. https://doi.org/10.3390/ijms24044039.
62. Dong K, Goyarts E, Rella A *et al.* Age associated decrease of MT-1 melatonin receptor in Human dermal skin fibroblasts impairs protection against UV-induced DNA damage. *Int J Mol Sci* 2020;**21**:326. https://doi.org/10.3390/ijms21010326.
63. Kleszczyński K, Tukaj S, Kruse N *et al.* Melatonin prevents ultraviolet radiation-induced alterations in plasma membrane potential and intracellular pH in human keratinocytes. *J Pineal Res* 2013;**54**:89–99. https://doi.org/10.1111/j.1600-079X.2012.01028.x.
64. Mansouri A, Gaou I, De Kerguenec C *et al.* An alcoholic binge causes massive degradation of hepatic mitochondrial DNA in mice. *Gastroenterology* 1999;**117**:181–90. https://doi.org/10.1016/s0016-5085(99)70566-4.
65. Díaz-Casado ME, Rusanova I, Aranda P *et al.* In vivo determination of mitochondrial respiration in 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-treated zebrafish reveals the efficacy of melatonin in restoring mitochondrial normalcy. *Zebrafish* 2018;**15**:15–26. https://doi.org/10.1089/zeb.2017.1479.
66. Pi H, Xu S, Reiter RJ *et al.* SIRT3-SOD2-mROS-dependent autophagy in cadmium-induced hepatotoxicity and salvage by melatonin. *Autophagy* 2015;**11**:1037–51. https://doi.org/10.1080/15548627.2015.1052208.
67. Slominski AT, Zmijewski MA, Semak I *et al.* Melatonin, mitochondria, and the skin. *Cell Mol Life Sci* 2017;**74**:3913–25. https://doi.org/10.1007/s00018-017-2617-7.
68. Venegas C, García JA, Escames G *et al.* Extrapineal melatonin: analysis of its subcellular distribution and daily fluctuations. *J Pineal Res* 2012;**52**:217–27. https://doi.org/10.1111/j.1600-079X.2011.00931.x.
69. Slominski AT, Semak I, Fischer TW *et al.* Metabolism of melatonin in the skin: why is it important? *Exp Dermatol* 2017;**26**:563–8. https://doi.org/10.1111/exd.13208.
70. Lu TX, Rothenberg ME.. MicroRNA. *J Allergy Clin Immunol* 2018;**141**:1202–7. https://doi.org/10.1016/j.jaci.2017.08.034.
71. Ahmed MI, Pickup ME, Rimmer AG *et al.* Interplay of MicroRNA-21 and SATB1 in epidermal keratinocytes during skin aging. *J Invest Dermatol* 2019;**139**:2538–42. https://doi.org/10.1016/j.jid.2019.04.022.
72. Srivastava A, Karlsson M, Marionnet C *et al.* Identification of chronological and photoageing-associated microRNAs in human skin. *Sci Rep* 2018;**8**:12990. https://doi.org/10.1038/s41598-018-31217-8.
73. Röck K, Tigges J, Sass S *et al.* miR-23a-3p causes cellular senescence by targeting hyaluronan synthase 2: possible implication for skin aging. *J Invest Dermatol* 2015;**135**:369–77. https://doi.org/10.1038/jid.2014.422.
74. Salminen A, Kaarniranta K, Kauppinen A. Photoaging: UV radiation-induced inflammation and immunosuppression accelerate the aging process in the skin. *Inflamm Res* 2022;**71**:817–31. https://doi.org/10.1007/s00011-022-01598-8.
75. Lee YI, Choi S, Roh WS *et al.* Cellular senescence and inflammaging in the skin microenvironment. *Int J Mol Sci* 2021;**22**:3849. https://doi.org/10.3390/ijms22083849.
76. Pilkington SM, Bulfone-Paus S, Griffiths CEM *et al.* Inflammaging and the skin. *J Invest Dermatol* 2021;**141**:1087–95. https://doi.org/10.1016/j.jid.2020.11.006.
77. Pilkington SM, Ogden S, Eaton LH *et al.* Lower levels of interleukin-1beta gene expression are associated with impaired Langerhans' cell migration in aged human skin. *Immunology* 2018;**153**:60–70. https://doi.org/10.1111/imm.12810.
78. Wang Y, Liu S, Li L *et al.* Peritoneal M2 macrophage-derived extracellular vesicles as natural multitarget nanotherapeutics to attenuate cytokine storms after severe infections. *J Control Release* 2022;**349**:118–32. https://doi.org/10.1016/j.jconrel.2022.06.063.
79. Gather L, Nath N, Falckenhayn C *et al.* Macrophages are polarized toward an inflammatory phenotype by their aged microenvironment in the Human skin. *J Invest Dermatol* 2022;**142**:3136–45. https://doi.org/10.1016/j.jid.2022.06.023.
80. Zhou Z, Liu J, Fu T *et al.* Phosphorylation regulates the binding of autophagy receptors to FIP200 Claw domain for selective autophagy initiation. *Nat Commun* 2021;**12**:1570. https://doi.org/10.1038/s41467-021-21874-1.
81. Yin Z, Liu X, Ariosa A *et al.* Psp2, a novel regulator of autophagy that promotes autophagy-related protein translation. *Cell Res* 2019;**29**:994–1008. https://doi.org/10.1038/s41422-019-0246-4.
82. Umar SA, Shahid NH, Nazir LA *et al.* Pharmacological activation of autophagy restores cellular homeostasis in ultraviolet-(B)-

induced skin photodamage. *Front Oncol* 2021;**11**:726066. https://doi.org/10.3389/fonc.2021.726066.
83. Chen Q, Zhang H, Yang Y *et al.* Metformin attenuates UVA-induced skin photoaging by suppressing mitophagy and the PI3K/AKT/mTOR pathway. *Int J Mol Sci* 2022;**23**:6960. https://doi.org/10.3390/ijms23136960.
84. Khan A, Ikram M, Muhammad T *et al.* Caffeine modulates cadmium-induced oxidative stress, neuroinflammation, and cognitive impairments by regulating nrf-2/HO-1 In vivo and In vitro. *J Clin Med* 2019;**8**:680. https://doi.org/10.3390/jcm8050680.
85. Li Y-F, Ouyang S-H, Tu L-F *et al.* Caffeine protects skin from oxidative stress-induced senescence through the activation of autophagy. *Theranostics* 2018;**8**:5713–30. https://doi.org/10.7150/thno.28778.
86. Kou X, Xu X, Chen C *et al.* The Fas/Fap-1/Cav-1 complex regulates IL-1RA secretion in mesenchymal stem cells to accelerate wound healing. *Sci Transl Med* 2018;**10**:432. https://doi.org/10.1126/scitranslmed.aai8524.
87. Zhou H, He Y, Xiong W *et al.* MSC based gene delivery methods and strategies improve the therapeutic efficacy of neurological diseases. *Bioact Mater* 2023;**23**:409–37. https://doi.org/10.1016/j.bioactmat.2022.11.007.
88. Lv F-J, Tuan RS, Cheung KMC *et al.* Concise review: the surface markers and identity of human mesenchymal stem cells. *Stem Cells* 2014;**32**:1408–19. https://doi.org/10.1002/stem.1681.
89. Kalluri R, Lebleu VS.. The biology, function, and biomedical applications of exosomes. *Science* 2020;**367**:eaau6977. https://doi.org/10.1126/science.aau6977.
90. Trams EG, Lauter CJ, Norman Salem J *et al.* Exfoliation of membrane ecto-enzymes in the form of micro-vesicles. *Biochim Biophys Acta* 1981;**645**:63–70. https://doi.org/10.1016/0005-2736(81)90512-5.
91. Meng W, Hao Y, He C *et al.* Exosome-orchestrated hypoxic tumor microenvironment. *Mol Cancer* 2019;**18**:57. https://doi.org/10.1186/s12943-019-0982-6.
92. Wang X, Wu R, Zhai P *et al.* Hypoxia promotes EV secretion by impairing lysosomal homeostasis in HNSCC through negative regulation of ATP6V1A by HIF-1α. *J Extracell Vesicles* 2023;**12**:e12310. https://doi.org/10.1002/jev2.12310.
93. Meldolesi J. Exosomes and ectosomes in intercellular communication. *Current biology: CB* 2018;**28**: R435–44. https://doi.org/10.1016/j.cub.2018.01.059.
94. Liang Y, Duan L, Lu J *et al.* Engineering exosomes for targeted drug delivery. *Theranostics* 2021;**11**:3183–95. https://doi.org/10.7150/thno.52570.
95. Liang G, Zhu Y, Ali DJ *et al.* Engineered exosomes for targeted co-delivery of miR-21 inhibitor and chemotherapeutics to reverse drug resistance in colon cancer. *J Nanobiotechnology* 2020;**18**:10. https://doi.org/10.1186/s12951-019-0563-2.
96. Yeo RWY, Lai RC, Zhang B *et al.* Mesenchymal stem cell: an efficient mass producer of exosomes for drug delivery. *Adv Drug Deliv Rev* 2013;**65**:336–41. https://doi.org/10.1016/j.addr.2012.07.001.
97. Jadczyk T, Faulkner A, Madeddu P. Stem cell therapy for cardiovascular disease: the demise of alchemy and rise of pharmacology. *Br J Pharmacol* 2013;**169**:247–68. https://doi.org/10.1111/j.1476-5381.2012.01965.x.
98. Chan AML, Sampasivam Y, Lokanathan Y. Biodistribution of mesenchymal stem cells (MSCs) in animal models and implied role of exosomes following systemic delivery of MSCs: a systematic review. *Am J Transl Res* 2022;**14**:2147–61.
99. Furlani D, Ugurlucan M, Ong L *et al.* Is the intravascular administration of mesenchymal stem cells safe? Mesenchymal stem cells and intravital microscopy. *Microvasc Res* 2009;**77**:370–6. https://doi.org/10.1016/j.mvr.2009.02.001.
100. Merino-González C, Zuñiga FA, Escudero C *et al.* Mesenchymal stem cell-derived extracellular vesicles promote angiogenesis: potencial clinical application. *Front Physiol* 2016;**7**:24. https://doi.org/10.3389/fphys.2016.00024.
101. Santos AC, Morais F, Simões A *et al.* Nanotechnology for the development of new cosmetic formulations. *Expert Opin Drug Deliv* 2019;**16**:313–30. https://doi.org/10.1080/17425247.2019.1585426.
102. Kaul S, Gulati N, Verma D *et al.* Role of nanotechnology in cosmeceuticals: A review of recent advances. *J Pharm (Cairo)* 2018;**2018**:3420204. https://doi.org/10.1155/2018/3420204.
103. Aziz ZAA, Mohd-Nasir H, Ahmad A *et al.* Role of nanotechnology for design and development of Cosmeceutical: application in makeup and skin care. *Front Chem* 2019;**7**:739. https://doi.org/10.3389/fchem.2019.00739.
104. Sharma A, Madhunapantula SV, Robertson GP. Toxicological considerations when creating nanoparticle-based drugs and drug delivery systems. *Expert Opin Drug Metab Toxicol* 2012;**8**:47–69. https://doi.org/10.1517/17425255.2012.637916.
105. Guo S-C, Tao S-C, Yin W-J *et al.* Exosomes derived from platelet-rich plasma promote the re-epithelization of chronic cutaneous wounds via activation of YAP in a diabetic rat model. *Theranostics* 2017;**7**:81–96. https://doi.org/10.7150/thno.16803.
106. Chi B, Zou A, Mao L *et al.* Empagliflozin-pretreated mesenchymal stem cell-derived small extracellular vesicles attenuated heart injury. *Oxid Med Cell Longev* 2023;**2023**:7747727. https://doi.org/10.1155/2023/7747727.
107. Magny R, Regazzetti A, Kessal K *et al.* Lipid annotation by combination of UHPLC-HRMS (MS), molecular networking, and retention time prediction: application to a lipidomic study of In vitro models of dry eye disease. *Metabolites* 2020;**10**: 225. https://doi.org/10.3390/metabo10060225.
108. Kong L, Zhang D, Huang S *et al.* Extracellular vesicles in mental disorders: A state-of-art review. *Int J Biol Sci* 2023;**19**:1094–109. https://doi.org/10.7150/ijbs.79666.
109. Yin X, Jiang L-H. Extracellular vesicles: targeting the heart. *Front Cardiovasc Med* 2022;**9**:1041481. https://doi.org/10.3389/fcvm.2022.1041481.
110. Lőrincz ÁM, Timár CI, Marosvári KA *et al.* Effect of storage on physical and functional properties of extracellular vesicles derived from neutrophilic granulocytes. *J Extracell Vesicles* 2014;**3**:25465. https://doi.org/10.3402/jev.v3.25465.
111. Zhou H, Yuen PST, Pisitkun T *et al.* Collection, storage, preservation, and normalization of human urinary exosomes for biomarker discovery. *Kidney Int* 2006;**69**:1471–6. https://doi.org/10.1038/sj.ki.5000273.
112. Mahanty S, Dakappa SS, Shariff R *et al.* Keratinocyte differentiation promotes ER stress-dependent lysosome biogenesis. *Cell Death Dis* 2019;**10**:269. https://doi.org/10.1038/s41419-019-1478-4.
113. Lee S, Choi YJ, Lee S *et al.* Protective effects of withagenin A diglucoside from indian Ginseng (Withania somnifera) against Human dermal fibroblast damaged by TNF-α stimulation. *Antioxidants (Basel, Switzerland)* 2022;**11**:2248. https://doi.org/10.3390/antiox11112248.
114. Zou Z, Long X, Zhao Q *et al.* A single-cell transcriptomic atlas of Human skin aging. *Dev Cell* 2021;**56**:383–97. https://doi.org/10.1016/j.devcel.2020.11.002.

115. Chang H, Chen J, Ding K *et al.* Highly-expressed lncRNA FOXD2-AS1 in adipose mesenchymal stem cell derived exosomes affects HaCaT cells via regulating miR-185-5p/ROCK2 axis. *Adipocyte* 2023;**12**:2173513. https://doi.org/10.1080/21623945.2023.2173513.
116. Zhao G, Liu F, Liu Z *et al.* MSC-derived exosomes attenuate cell death through suppressing AIF nucleus translocation and enhance cutaneous wound healing. *Stem Cell Res Ther* 2020;**11**:174. https://doi.org/10.1186/s13287-020-01616-8.
117. Zhang W, Bai X, Zhao B *et al.* Cell-free therapy based on adipose tissue stem cell-derived exosomes promotes wound healing via the PI3K/Akt signaling pathway. *Exp Cell Res* 2018;**370**:333–42. https://doi.org/10.1016/j.yexcr.2018.06.035.
118. Jiang T, Wang Z, Sun J. Human bone marrow mesenchymal stem cell-derived exosomes stimulate cutaneous wound healing mediates through TGF-β/smad signaling pathway. *Stem Cell Res Ther* 2020;**11**:198. https://doi.org/10.1186/s13287-020-01723-6.
119. Cui N, Hu M, Khalil RA. Biochemical and biological attributes of matrix metalloproteinases. *Prog Mol Biol Transl Sci* 2017;**147**:1–73. https://doi.org/10.1016/bs.pmbts.2017.02.005.
120. Szilágyi Z, Németh Z, Bakos J *et al.* Assessment of inflammation in 3D reconstructed Human skin exposed to combined exposure to ultraviolet and Wi-fi radiation. *Int J Mol Sci* 2023;**24**:2853. https://doi.org/10.3390/ijms24032853.
121. Kim H, Jang J, Song MJ *et al.* Inhibition of matrix metalloproteinase expression by selective clearing of senescent dermal fibroblasts attenuates ultraviolet-induced photoaging. *Biomed Pharmacother* 2022;**150**:113034. https://doi.org/10.1016/j.biopha.2022.113034.
122. Park J-Y, Lee JY, Kim Y *et al.* Latilactobacillus sakei Wikim0066 protects skin through MMP regulation on UVB-irradiated In vitro and In vivo model. *Nutrients* 2023;**15**:726. https://doi.org/10.3390/nu15030726.
123. Deng M, Yu TZ, Li D *et al.* Human umbilical cord mesenchymal stem cell-derived and dermal fibroblast-derived extracellular vesicles protect dermal fibroblasts from ultraviolet radiation-induced photoaging in vitro. *Photochem Photobiol Sci* 2020;**19**:406–14. https://doi.org/10.1039/c9pp00421a.
124. Gao W, Yuan L-M, Zhang Y *et al.* miR-1246-overexpressing exosomes suppress UVB-induced photoaging via regulation of TGF-β/smad and attenuation of MAPK/AP-1 pathway. *Photochem Photobiol Sci* 2023;**22**:135–46. https://doi.org/10.1007/s43630-022-00304-1.
125. Zhang Y, Zhang M, Yao A *et al.* Circ_0011129 encapsulated by the small extracellular vesicles derived from Human stem cells ameliorate skin photoaging. *Int J Mol Sci* 2022;**23**:15390. https://doi.org/10.3390/ijms232315390.
126. Yen C-A, Curran SP.. Incomplete proline catabolism drives premature sperm aging. *Aging Cell* 2021;**20**: e13308. https://doi.org/10.1111/acel.13308.
127. Liu X, Xing Y, Yuen M *et al.* Anti-aging effect and mechanism of proanthocyanidins extracted from sea buckthorn on hydrogen peroxide-induced aging Human skin fibroblasts. *Antioxidants (Basel, Switzerland)* 2022;**11**:1900. https://doi.org/10.3390/antiox11101900.
128. Feng W, Han X, Hu H *et al.* 2D vanadium carbide MXenzyme to alleviate ROS-mediated inflammatory and neurodegenerative diseases. *Nat Commun* 2021;**12**:2203. https://doi.org/10.1038/s41467-021-22278-x.
129. Jiang Q, Yin J, Chen J *et al.* Mitochondria-targeted antioxidants: A step towards disease treatment. *Oxid Med Cell Longev* 2020;**2020**:8837893. https://doi.org/10.1155/2020/8837893.
130. Wang T, Jian Z, Baskys A *et al.* MSC-derived exosomes protect against oxidative stress-induced skin injury via adaptive regulation of the NRF2 defense system. *Biomaterials* 2020;**257**:120264. https://doi.org/10.1016/j.biomaterials.2020.120264.
131. Shiekh PA, Singh A, Kumar A. Exosome laden oxygen releasing antioxidant and antibacterial cryogel wound dressing OxOBand alleviate diabetic and infectious wound healing. *Biomaterials* 2020;**249**:120020. https://doi.org/10.1016/j.biomaterials.2020.120020.
132. Li X, Xie X, Lian W *et al.* Exosomes from adipose-derived stem cells overexpressing Nrf2 accelerate cutaneous wound healing by promoting vascularization in a diabetic foot ulcer rat model. *Exp Mol Med* 2018;**50**:1–14. https://doi.org/10.1038/s12276-018-0058-5.
133. Wu P, Zhang B, Han X *et al.* HucMSC exosome-delivered 14-3-3ζ alleviates ultraviolet radiation-induced photodamage via SIRT1 pathway modulation. *Aging* 2021;**13**:11542–63. https://doi.org/10.18632/aging.202851.
134. Miró L, Garcia-Just A, Amat C *et al.* Dietary animal plasma proteins improve the intestinal immune response in senescent mice. *Nutrients* 2017;**9**:1346. https://doi.org/10.3390/nu9121346.
135. Qin Z–Y, Gu X, Chen Y–L *et al.* Toll–like receptor 4 activates the NLRP3 inflammasome pathway and periodontal inflammaging by inhibiting bmi–1 expression. *Int J Mol Med* 2021;**47**:137–50. https://doi.org/10.3892/ijmm.2020.4787.
136. Agrawal A, Agrawal S, Gupta S. Role of dendritic cells in inflammation and loss of tolerance in the elderly. *Front Immunol* 2017;**8**:896. https://doi.org/10.3389/fimmu.2017.00896.
137. Rodrigues LP, Teixeira VR, Alencar-Silva T *et al.* Hallmarks of aging and immunosenescence: connecting the dots. *Cytokine Growth Factor Rev* 2021;**59**:9–21. https://doi.org/10.1016/j.cytogfr.2021.01.006.
138. Pilkington SM, Bulfone-Paus S, Griffiths CEM *et al.* Inflammaging and the skin. *J Invest Dermatol* 2021;**141**:1087–95. https://doi.org/10.1016/j.jid.2020.11.006.
139. Hasegawa T, Nakashima M, Suzuki Y. Nuclear DNA damage-triggered NLRP3 inflammasome activation promotes UVB-induced inflammatory responses in human keratinocytes. *Biochem Biophys Res Commun* 2016;**477**:329–35. https://doi.org/10.1016/j.bbrc.2016.06.106.
140. Xiao T, Chen Y, Song C *et al.* Possible treatment for UVB-induced skin injury: anti-inflammatory and cytoprotective role of metformin in UVB-irradiated keratinocytes. *J Dermatol Sci* 2021;**102**:25–35, https://doi.org/10.1016/j.jdermsci.2021.02.002.
141. Shi Y, Wang Y, Li Q *et al.* Immunoregulatory mechanisms of mesenchymal stem and stromal cells in inflammatory diseases. *Nat Rev Nephrol* 2018;**14**:493–507. https://doi.org/10.1038/s41581-018-0023-5.
142. Zhang B, Lai RC, Sim WK *et al.* Topical application of mesenchymal stem cell exosomes alleviates the imiquimod induced psoriasis-like inflammation. *Int J Mol Sci* 2021;**22**:720. https://doi.org/10.3390/ijms22020720.
143. Cho BS, Kim JO, Ha DH *et al.* Exosomes derived from human adipose tissue-derived mesenchymal stem cells alleviate atopic

dermatitis. *Stem Cell Res Ther* 2018;**9**:187. https://doi.org/10.1186/s13287-018-0939-5.

144. Patel RS, Impreso S, Lui A *et al.* Long noncoding RNA GAS5 contained in exosomes derived from Human adipose stem cells promotes repair and modulates inflammation in a chronic dermal wound healing model. *Biology* 2022;**11**:426. https://doi.org/10.3390/biology11030426.
145. Liu W, Yu M, Xie D *et al.* Melatonin-stimulated MSC-derived exosomes improve diabetic wound healing through regulating macrophage M1 and M2 polarization by targeting the PTEN/AKT pathway. *Stem Cell Res Ther* 2020;**11**:259. https://doi.org/10.1186/s13287-020-01756-x.
146. Heo JS. Selenium-stimulated exosomes enhance wound healing by modulating inflammation and angiogenesis. *Int J Mol Sci* 2022;**23**:11543. https://doi.org/10.3390/ijms231911543.
147. Park G-H, Kwon HH, Seok J *et al.* Efficacy of combined treatment with human adipose tissue stem cell-derived exosome-containing solution and microneedling for facial skin aging: A 12-week prospective, randomized, split-face study. *J Cosmet Dermatol* 2023;**22**:3418–26. https://doi.org/10.1111/jocd.15872.
148. Rohde E, Pachler K, Gimona M. Manufacturing and characterization of extracellular vesicles from umbilical cord-derived mesenchymal stromal cells for clinical testing. *Cytotherapy* 2019;**21**:581–92. https://doi.org/10.1016/j.jcyt.2018.12.006.
149. Imai T, Takahashi Y, Nishikawa M *et al.* Macrophage-dependent clearance of systemically administered B16BL6-derived exosomes from the blood circulation in mice. *J Extracell Vesicles* 2015;**4**:26238. https://doi.org/10.3402/jev.v4.26238.
150. Cao H, Duan L, Zhang Y *et al.* Current hydrogel advances in physicochemical and biological response-driven biomedical application diversity. *Signal Transduct Target Ther* 2021;**6**:426. https://doi.org/10.1038/s41392-021-00830-x.
151. Lai W-F, He Z-D. Design and fabrication of hydrogel-based nanoparticulate systems for in vivo drug delivery. *J Control Release* 2016;**243**:269–82. https://doi.org/10.1016/j.jconrel.2016.10.013.
152. Teixeira MO, Antunes JC, Felgueiras HP. Recent advances in Fiber-hydrogel composites for wound healing and drug delivery systems. *Antibiotics (Basel)* 2021;**10**:248. https://doi.org/10.3390/antibiotics10030248.
153. Wang Q, Qu Y, Zhang Z *et al.* Injectable DNA hydrogel-based local drug delivery and immunotherapy. *Gels* 2022;**8**:400. https://doi.org/10.3390/gels8070400.
154. Mol EA, Lei Z, Roefs MT *et al.* Injectable supramolecular ureidopyrimidinone hydrogels provide sustained release of extracellular vesicle therapeutics. *Adv Healthc Mater* 2019;**8**: e1900847. https://doi.org/10.1002/adhm.201900847.
155. Zhang K, Chen X, Li H *et al.* A nitric oxide-releasing hydrogel for enhancing the therapeutic effects of mesenchymal stem cell therapy for hindlimb ischemia. *Acta Biomater* 2020;**113**:289–304. https://doi.org/10.1016/j.actbio.2020.07.011.
156. Zhang S, Liu Y, Zhang X *et al.* Prostaglandin E(2) hydrogel improves cutaneous wound healing via M2 macrophages polarization. *Theranostics* 2018;**8**:5348–61. https://doi.org/10.7150/thno.27385.
157. Zhao X, Liu Y, Jia P *et al.* Chitosan hydrogel-loaded MSC-derived extracellular vesicles promote skin rejuvenation by ameliorating the senescence of dermal fibroblasts. *Stem Cell Res Ther* 2021;**12**:196. https://doi.org/10.1186/s13287-021-02262-4.
158. You DG, An JY, Um W *et al.* Stem cell-derived extracellular vesicle-bearing dermal filler ameliorates the dermis microenvironment by supporting CD301b-expressing macrophages. *ACS Nano* 2022;**16**:251–60. https://doi.org/10.1021/acsnano.1c06096.
159. Ma G, Wu C.. Microneedle, bio-microneedle and bio-inspired microneedle: A review. *J Control Release* 2017;**251**:11–23. https://doi.org/10.1016/j.jconrel.2017.02.011.
160. Hong JY, Kwon T-R, Kim JH *et al.* Prospective, preclinical comparison of the performance between radiofrequency microneedling and microneedling alone in reversing photoaged skin. *J Cosmet Dermatol* 2020;**19**:1105–9. https://doi.org/10.1111/jocd.13116.
161. Cao Z, Jin S, Wang P *et al.* Microneedle based adipose derived stem cells-derived extracellular vesicles therapy ameliorates UV-induced photoaging in SKH-1 mice. *J Biomed Mater Res A* 2021;**109**:1849–57. https://doi.org/10.1002/jbm.a.37177.
162. You Y, Tian Y, Yang Z *et al.* Intradermally delivered mRNA-encapsulating extracellular vesicles for collagen-replacement therapy. *Nat Biomed Eng* 2023;**7**:887–900. https://doi.org/10.1038/s41551-022-00989-w.
163. Zhang S, Ou H, Liu C *et al.* Skin delivery of hydrophilic biomacromolecules using marine sponge spicules. *Mol Pharm* 2017;**14**:3188–200. https://doi.org/10.1021/acs.molpharmaceut.7b00468.
164. Zhang K, Yu L, Li F-R *et al.* Topical application of exosomes derived from Human umbilical cord mesenchymal stem cells in combination with sponge spicules for treatment of photoaging. *Int J Nanomedicine* 2020;**15**:2859–72. https://doi.org/10.2147/IJN.S249751.
165. Nkosi D, Howell LA, Cheerathodi MR *et al.* Transmembrane domains mediate intra- and extracellular trafficking of Epstein-Barr Virus latent membrane protein 1. *J Virol* 2018;**92**:e00280–18. https://doi.org/10.1128/jvi.00280-18.
166. Fang Y, Wu N, Gan X *et al.* Higher-order oligomerization targets plasma membrane proteins and HIV gag to exosomes. *PLoS Biol* 2007;**5**: e158. https://doi.org/10.1371/journal.pbio.0050158.
167. Gan X, Gould SJ. Identification of an inhibitory budding signal that blocks the release of HIV particles and exosome/microvesicle proteins. *Mol Biol Cell* 2011;**22**:817–30. https://doi.org/10.1091/mbc.E10-07-0625.
168. Hu CMJ, Zhang L. Nanoparticle-based combination therapy toward overcoming drug resistance in cancer. *Biochem Pharmacol* 2012;**83**:1104–11. https://doi.org/10.1016/j.bcp.2012.01.008.
169. Gibis M, Rahn N, Weiss J. Physical and oxidative stability of uncoated and chitosan-coated liposomes containing grape seed extract. *Pharmaceutics* 2013;**5**:421–33. https://doi.org/10.3390/pharmaceutics5030421.
170. Hu CMJ, Zhang L, Aryal S *et al.* Erythrocyte membrane-camouflaged polymeric nanoparticles as a biomimetic delivery platform. *Proc Natl Acad Sci USA* 2011;**108**:10980–5. https://doi.org/10.1073/pnas.1106634108.
171. Hu CMJ, Fang RH, Wang K-C *et al.* Nanoparticle biointerfacing by platelet membrane cloaking. *Nature* 2015;**526**:118–21. https://doi.org/10.1038/nature15373.
172. Boada C, Zinger A, Tsao C *et al.* Rapamycin-loaded biomimetic nanoparticles reverse vascular inflammation. *Circ Res* 2020;**126**:25–37. https://doi.org/10.1161/CIRCRESAHA.119.315185. Epub 2019 Oct 24. Erratum in: *Circ Res*. 2020 Jul 3;**127**(2):e77. PMID: 31647755.
173. Evers MJW, Van De Wakker SI, De Groot EM *et al.* Functional siRNA delivery by extracellular vesicle-liposome hybrid nanoparticles. *Adv Healthc Mater* 2022;**11**: e2101202. https://doi.org/10.1002/adhm.202101202.
174. Zhou X, Miao Y, Wang Y *et al.* Tumour-derived extracellular vesicle membrane hybrid lipid nanovesicles enhance siRNA

delivery by tumour-homing and intracellular freeway transportation. *J Extracell Vesicles* 2022;**11**: e12198. https://doi.org/10.1002/jev2.12198.
175. Belhadj Z, He B, Deng H *et al.* A combined "eat me/don't eat me" strategy based on extracellular vesicles for anticancer nanomedicine. *J Extracell Vesicles* 2020;**9**:1806444. https://doi.org/10.1080/20013078.2020.1806444.
176. Delcayre A, Estelles A, Sperinde J *et al.* Exosome display technology: applications to the development of new diagnostics and therapeutics. *Blood Cells Mol Dis* 2005;**35**:158–68. https://doi.org/10.1016/j.bcmd.2005.07.003.
177. Hartman ZC, Wei J, Glass OK *et al.* Increasing vaccine potency through exosome antigen targeting. *Vaccine* 2011;**29**:9361–7. https://doi.org/10.1016/j.vaccine.2011.09.133.
178. Wang J-H, Forterre AV, Zhao J *et al.* Anti-HER2 scFv-directed extracellular vesicle-mediated mRNA-based gene delivery inhibits growth of HER2-positive Human breast tumor xenografts by prodrug activation. *Mol Cancer Ther* 2018;**17**:1133–42. https://doi.org/10.1158/1535-7163.Mct-17-0827.
179. Longatti A, Schindler C, Collinson A *et al.* High affinity single-chain variable fragments are specific and versatile targeting motifs for extracellular vesicles. *Nanoscale* 2018;**10**:14230–44. https://doi.org/10.1039/c8nr03970d.
180. Alvarez-Erviti L, Seow Y, Yin H *et al.* Delivery of siRNA to the mouse brain by systemic injection of targeted exosomes. *Nat Biotechnol* 2011;**29**:341–5. https://doi.org/10.1038/nbt.1807.
181. Antes TJ, Middleton RC, Luther KM *et al.* Targeting extracellular vesicles to injured tissue using membrane cloaking and surface display. *J Nanobiotechnology* 2018;**16**:61. https://doi.org/10.1186/s12951-018-0388-4.
182. Cao Y, Wu T, Zhang K *et al.* Engineered exosome-mediated Near-Infrared-II region V(2)C quantum dot delivery for nucleus-target low-temperature photothermal therapy. *ACS Nano* 2019;**13**:1499–510. https://doi.org/10.1021/acsnano.8b07224.
183. Qi H, Liu C, Long L *et al.* Blood exosomes endowed with magnetic and targeting properties for cancer therapy. *ACS Nano* 2016;**10**:3323–33. https://doi.org/10.1021/acsnano.5b06939.
184. Vyas KS, Kaufman J, Munavalli GS *et al.* Exosomes: the latest in regenerative aesthetics. *Regen Med* 2023;**18**:181–94. https://doi.org/10.2217/rme-2022-0134.
185. Wu H, Zhang Z, Zhang Y *et al.* Extracellular vesicle: A magic lamp to treat skin aging, refractory wound, and pigmented dermatosis? *Front Bioeng Biotechnol* 2022;**10**:1043320. https://doi.org/10.3389/fbioe.2022.1043320.
186. Kamerkar S, Lebleu VS, Sugimoto H *et al.* Exosomes facilitate therapeutic targeting of oncogenic KRAS in pancreatic cancer. *Nature* 2017;**546**:498–503. https://doi.org/10.1038/nature22341.
187. Yang Z, Shi J, Xie J *et al.* Large-scale generation of functional mRNA-encapsulating exosomes via cellular nanoporation. *Nat Biomed Eng* 2020;**4**:69–83. https://doi.org/10.1038/s41551-019-0485-1.
188. Ravi A, Sadhna D, Nagpaal D *et al.* Needle free injection technology: A complete insight. *Int J Pharm Investig* 2015;**5**:192–9. https://doi.org/10.4103/2230-973x.167662.
189. Hu S, Li Z, Cores J *et al.* Needle-free injection of exosomes derived from human dermal fibroblast spheroids ameliorates skin photoaging. *ACS Nano* 2019;**13**:11273–82. https://doi.org/10.1021/acsnano.9b04384.
190. Zheng G, Huang R, Qiu G *et al.* Mesenchymal stromal cell-derived extracellular vesicles: regenerative and immunomodulatory effects and potential applications in sepsis. *Cell Tissue Res* 2018;**374**:1–15. https://doi.org/10.1007/s00441-018-2871-5.
191. Cully M. Exosome-based candidates move into the clinic. *Nat Rev Drug Discov* 2021;**20**:6–7. https://doi.org/10.1038/d41573-020-00220-y.
192. Görgens A, Corso G, Hagey DW *et al.* Identification of storage conditions stabilizing extracellular vesicles preparations. *J Extracell Vesicles* 2022;**11**: e12238. https://doi.org/10.1002/jev2.12238.
193. Zhang K, Cheng K. Stem cell-derived exosome versus stem cell therapy. *Nat Rev Bioeng* 2023;**12**:1–2. https://doi.org/10.1038/s44222-023-00064-2.
194. Elsharkasy OM, Nordin JZ, Hagey DW *et al.* Extracellular vesicles as drug delivery systems: why and how? *Adv Drug Deliv Rev* 2020;**159**:332–43. https://doi.org/10.1016/j.addr.2020.04.004.
195. Meng W, He C, Hao Y *et al.* Prospects and challenges of extracellular vesicle-based drug delivery system: considering cell source. *Drug Deliv* 2020;**27**:585–98. https://doi.org/10.1080/10717544.2020.1748758.
196. Ng K. Penetration enhancement of topical formulations. *Pharmaceutics* 2018;**10**:51. https://doi.org/10.3390/pharmaceutics10020051.

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