# MCT EXOSOMES

## Unleashing the power of autologous exosomes

The possibility of generating,
concentrating and administering exosomes
and other EVs signals a new era exponentially
increasing the power of regenerative medicine
aesthetic treatments. **MCT EXOSOMES** goes
one step beyond cells, achieving autologous
injectable exosomes for medical use
and awesome aesthetic and clinical results.

# MCT EXOSOMES
## One step beyond cells

MCT is a medical system that can improve any autologous product, such as PRP or stem cells concentrates, and stimulates the exosome production and deliverance. Through photothermal energy it delivers extraordinary growth factor concentration and boosts cells, filling them with ATP and huge amounts of exosomes.

MCT autologous products stand out for their remarkably enhanced regenerative power, achieving spectacular skin rejuvenation, tissue regeneration and aesthetic and clinical results.

# MCT System

MCT System is the result of focused and dedicated R&D efforts, aimed at creating a device that could bring the benefits of photothermal stimulation at the doctor's office. Its results are impressive and its potential, breathtaking.

- Enhanced regenerative power
- Amazing amount of exosomes
- Autologous high-quality exosomes
- Extraordinary clinical and aesthetic results

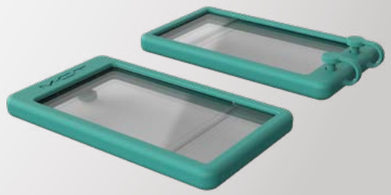


### MCT Unit®

MCT Unit® boosts existing regenerative medicine treatments simply and quickly. Through the application of electromagnetic and thermal energy, it improves the general characteristics of autologous products, such as growth factor content, ATP production, and cell or platelet performance. It can handle a wide range of temperatures and multiple wavelengths which are set up with user-friendly, one-touch presets.

### MCT Kit®

MCT Kit® is a patented device with a specific shape and chemical composition, exclusively developed for photothermal conditioning. It is made of a medical grade polymer that guarantees optimal scattering, transmittance, and other optical properties, ensuring that the emitted light will reach the target effectively. Its unique geometry grants an excellent surface/volume energy exposure ratio. MCT Kit® can allocate 10 mL of any autologous preparation.



# 3
easy steps for
## MCT EXOSOMES

**Step 01**

**Obtain autologous material**

**Step 02**

**Insert it into the MCT Kit®**

**Step 03**

**Set up the MCT Unit®**

# MCT EXOSOMES
Unleashing the power
of autologous exosomes

# MCT EXOSOMES

## Increased growth factors

Photothermal stimulation elicits controlled and sustained release of growth factors from platelets, achieving a more physiological effect.

## Autologous EXOSOME production

Cell and platelet photothermal conditioning stimulates exosome production and release, resulting in an autologous secretome with enhanced regenerative power.

## ATP Synthesis Stimulation

Photothermal enhancement boosting stimulates cellular metabolism and promotes ATP synthesis.

# EXOSOMES and blue light

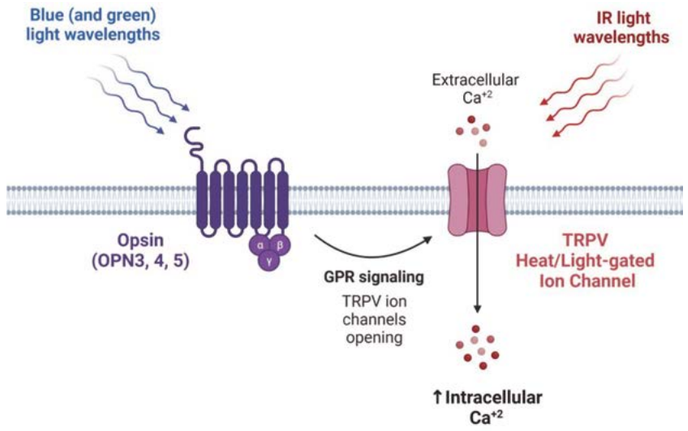
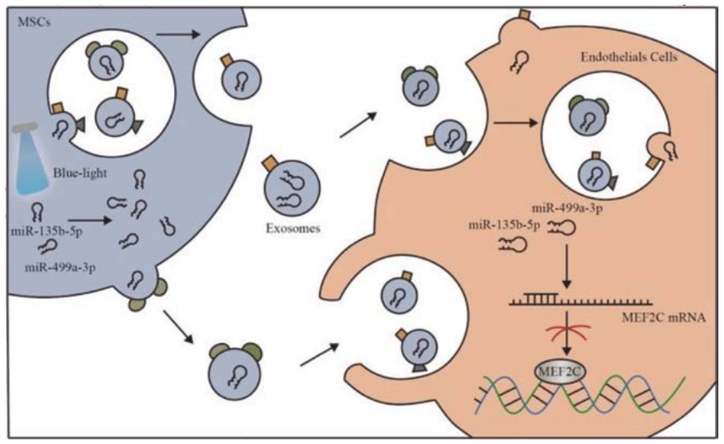


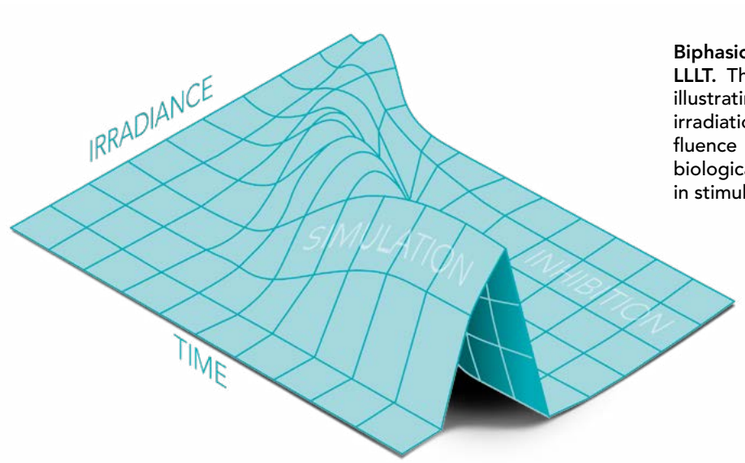

**Illustration of the possible involvement of heat/light-gated TRPV ion channels in PBM and IR Therapy.** Blue (or green) wavelengths could be absorbed by opsins based on photoisomerization of a retinal cofactor, leading to GPR signaling which opens TRPV ion channels. IR wavelengths could be absorbed by nanostructured water leading to a conformational change in the protein, which also opens TRPV ion channels. The influx of calcium affects mitochondria in the cells producing increased ATP and a brief burst of ROS. Eventually transcription factors are activated leading to long-lasting changes in the tissue. Adapted from: Sharma, S. K. et al. Role of opsins and light or heat activated transient receptor potential ion channels in the mechanisms of photo-biomodulation and infrared therapy. (2023). Journal of Photochemistry and Photobiology. Figure created with BioRender.com




**Putative mechanism by which blue light increases the two miRNAs to activate endothelial cells.** Extracted from: Yang K, et al. Exposure to blue light stimulates the proangiogenic capability of exosomes derived from human umbilical cord mesenchymal stem cells. (2019). Stem Cell Res Ther.

- MSCs constitutively express opsins for light responsiveness.
- Blue light-stimulation upregulates two miRNAs present in MSCs exosomes.
- These miRNAs enhance the proangiogenic potential by stimulating ECs.

# Total energy control



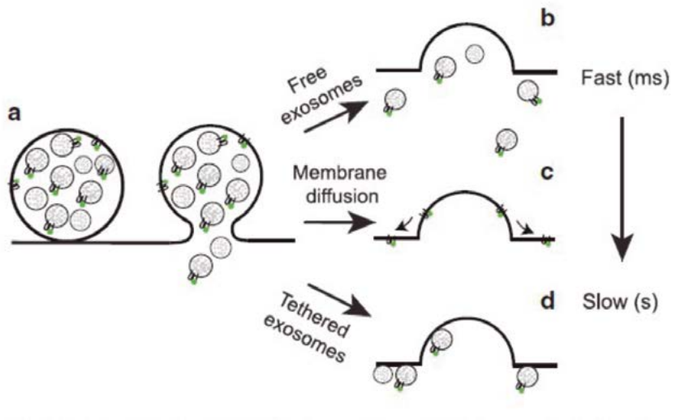

**Biphasic dose response in LLLT.** Three-dimensional plot illustrating effects of varying irradiation time equivalent to fluence or irradiance on the biological response resulting in stimulation or inhibition.

If the light applied is not of sufficient irradiance or the irradiation time is too short, then there will be no response. If the irradiance is too high or irradiation time is too long, then the response may be inhibited. Somewhere in between is the optimal combination of irradiance and time for stimulation.

Text and figure extracted from Chung, H. et al. The nuts and bolts of low-level laser (light) therapy. (2012). Ann Biomed Eng.

# EXOSOMES and temperature

- MVE fusion is a constitutive process enhanced in the presence of $Ca^{2+}$.
- MVE fusion is temperature dependent.
- Exosome content release is temperature dependent.
- Temperature affects docking and post-fusion events.



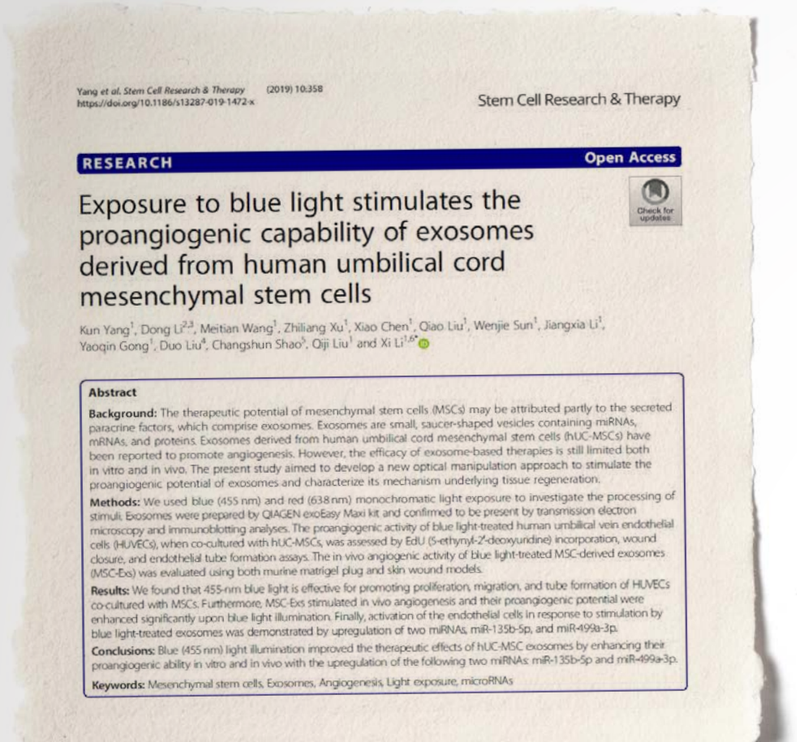

**Model of MVE fusion and how CD63 leaves the fusion site.** (A) MVEs first dock then fuse. Postfusion, three outcomes can occur simultaneously in single fusion events: (B) exosomes quickly leave the fusion site (24%, <0.5 s), (C) CD63-pHluorin on the endosomal membrane diffuses into the plasma membrane (36%, 1-2 s). or (D) tethered exosomes remain attached to the membrane and slowly leave the fusion site (40%, 5-10 s). The percent of each component and approximate time to leave the fusion site were determined from a simulation of the average fusion decay mesured at 37°C.

| | D (μm²/s) | 23°C | 27°C | 32°C | 37°C |
|---|---|---|---|---|---|
| **No. of events** | - | 86 | 83 | 77 | 98 |
| **$D_{tether}$ (μm²/s)** | varies | 0.0015 | 0.003 | 0.004 | 0.004 |
| **Fraction attached exosomes** | varies | 0.5 | 0.5 | 0.45 | 0.4 |
| **Fraction free exosomes** | 6.5 | 0.18 | 0.18 | 0.18 | 0.24 |
| **Fraction endosomal** | 0.039 | 0.32 | 0.32 | 0.37 | 0.36 |

Simulated to best match the average intensity loss trace at each temperature. $D_{tether}$, the fraction attached, and the fraction free were unconstrained. The fraction on the endosomal membrane was constrained to 0.3 ± 0.1.

Figure and table extracted from Mahmood, A. et al. Exosome secretion kinetics are controlled by temperature. (2023). Biophys J.

Photothermal conditioning stimulates exosome production and release from cells and platelets. They are pivotal for homeostasis, vascular integrity, inflammation regulation, and angiogenesis, directly impacting on health and aging.



Yang et al. Stem Cell Research & Therapy (2019) 10:358
https://doi.org/10.1186/s13287-019-1472-x

Stem Cell Research & Therapy

RESEARCH Open Access

# Exposure to blue light stimulates the proangiogenic capability of exosomes derived from human umbilical cord mesenchymal stem cells

Kun Yang[1], Dong Li[2,3], Meitian Wang[1], Zhiliang Xu[1], Xiao Chen[1], Qiao Liu[1], Wenjie Sun[1], Jiangxia Li[1], Yaoqin Gong[1], Duo Liu[4], Changshun Shao[5], Qiji Liu[1] and Xi Li[1,6*]

**Abstract**

**Background:** The therapeutic potential of mesenchymal stem cells (MSCs) may be attributed partly to the secreted paracrine factors, which comprise exosomes. Exosomes are small, saucer-shaped vesicles containing miRNAs, mRNAs, and proteins. Exosomes derived from human umbilical cord mesenchymal stem cells (hUC-MSCs) have been reported to promote angiogenesis. However, the efficacy of exosome-based therapies is still limited both in vitro and in vivo. The present study aimed to develop a new optical manipulation approach to stimulate the proangiogenic potential of exosomes and characterize its mechanism underlying tissue regeneration.

**Methods:** We used blue (455 nm) and red (638 nm) monochromatic light exposure to investigate the processing of stimuli. Exosomes were prepared by QIAGEN exoEasy Maxi kit and confirmed to be present by transmission electron microscopy and immunoblotting analyses. The proangiogenic activity of blue light-treated human umbilical vein endothelial cells (HUVECs), when co-cultured with hUC-MSCs, was assessed by EdU (5-ethynyl-2′-deoxyuridine) incorporation, wound closure, and endothelial tube formation assays. The in vivo angiogenic activity of blue light-treated MSC-derived exosomes (MSC-Exs) was evaluated using both murine matrigel plug and skin wound models.

**Results:** We found that 455-nm blue light is effective for promoting proliferation, migration, and tube formation of HUVECs co-cultured with MSCs. Furthermore, MSC-Exs stimulated in vivo angiogenesis and their proangiogenic potential were enhanced significantly upon blue light illumination. Finally, activation of the endothelial cells in response to stimulation by blue light-treated exosomes was demonstrated by upregulation of two miRNAs, miR-135b-5p, and miR-499a-3p.

**Conclusions:** Blue (455 nm) light illumination improved the therapeutic effects of hUC-MSC exosomes by enhancing their proangiogenic ability in vitro and in vivo with the upregulation of the following two miRNAs: miR-135b-5p and miR-499a-3p.

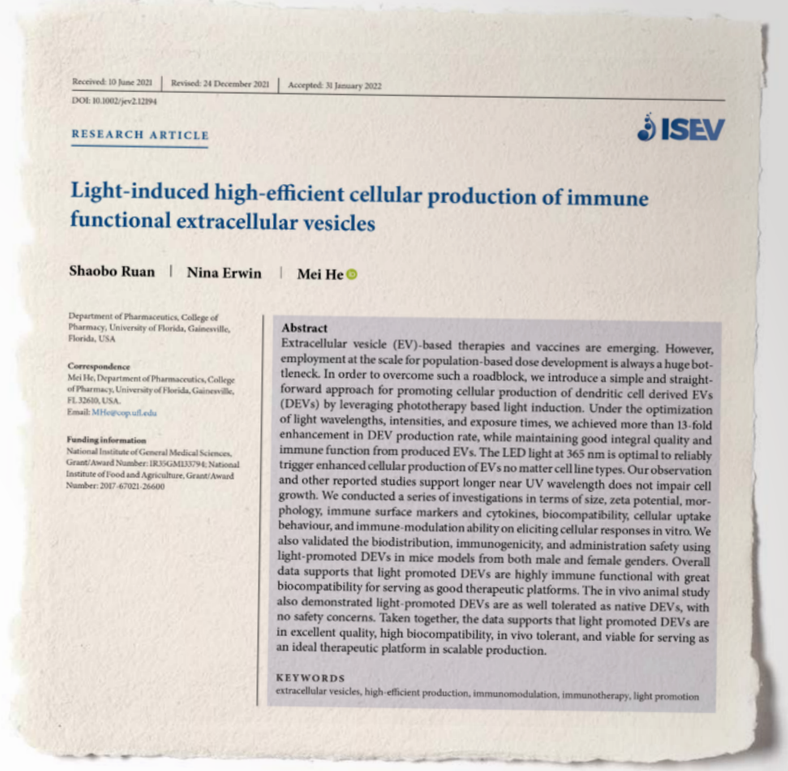
**Keywords:** Mesenchymal stem cells, Exosomes, Angiogenesis, Light exposure, microRNAs

## Highlights

- MSCs constitutionally express opsins for light responsiveness.
- Blue light promotes proangiogenic ability of MSC-Exosomes in vitro & in vivo.
- Blue light illumination of exosomes improves their therapeutic effects.

## Highlights

- Under the optimization of light wavelengths, intensities, and exposure times, more than 13-fold enhancement in DEV production rate is achieved, while maintaining integral quality and immune function from produced EVs.



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RESEARCH ARTICLE

ISEV

# Light-induced high-efficient cellular production of immune functional extracellular vesicles

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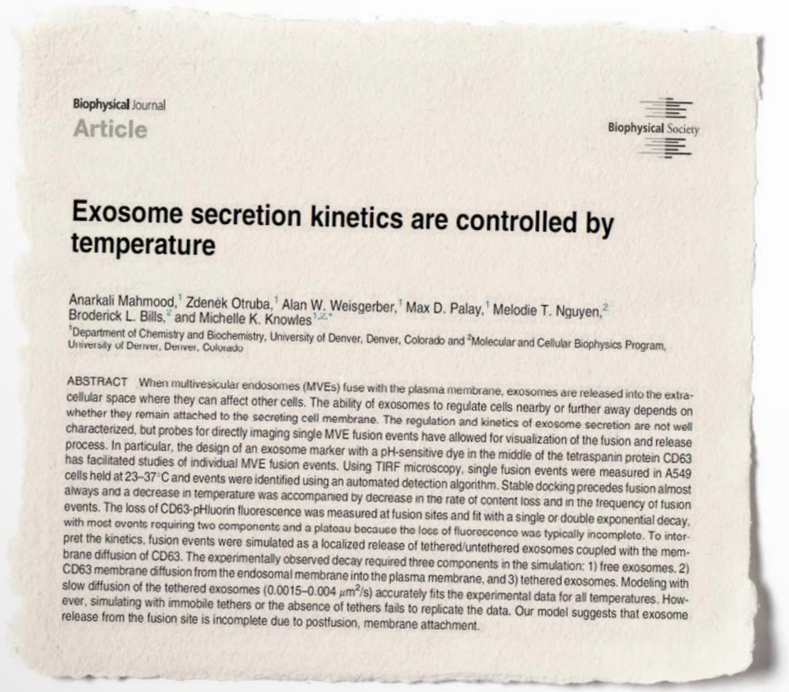
**Abstract**

Extracellular vesicle (EV)-based therapies and vaccines are emerging. However, employment at the scale for population-based dose development is always a huge bottleneck. In order to overcome such a roadblock, we introduce a simple and straightforward approach for promoting cellular production of dendritic cell derived EVs (DEVs) by leveraging phototherapy based light induction. Under the optimization of light wavelengths, intensities, and exposure times, we achieved more than 13-fold enhancement in DEV production rate, while maintaining good integral quality and immune function from produced EVs. The LED light at 365 nm is optimal to reliably trigger enhanced cellular production of EVs no matter cell line types. Our observation and other reported studies support longer near UV wavelength does not impair cell growth. We conducted a series of investigations in terms of size, zeta potential, morphology, immune surface markers and cytokines, biocompatibility, cellular uptake behaviour, and immune-modulation ability on eliciting cellular responses in vitro. We also validated the biodistribution, immunogenicity, and administration safety using light-promoted DEVs in mice models from both male and female genders. Overall data supports that light promoted DEVs are highly immune functional with great biocompatibility for serving as good therapeutic platforms. The in vivo animal study also demonstrated light-promoted DEVs are as well tolerated as native DEVs, with no safety concerns. Taken together, the data supports that light promoted DEVs are in excellent quality, high biocompatibility, in vivo tolerant, and viable for serving as an ideal therapeutic platform in scalable production.

KEYWORDS
extracellular vesicles, high-efficient production, immunomodulation, immunotherapy, light promotion

## Highlights

- MVE fusion is a constitutive process enhanced in the presence of $Ca^{2+}$.
- Exosomes are secreted from MVEs more frequently at higher temperatures.
- Exosome content release is temperature dependent.
- Temperature affects docking and post-fusion events.



Biophysical Journal
Article

Biophysical Society

# Exosome secretion kinetics are controlled by temperature

Anarkali Mahmood,[1] Zdeněk Otruba,[1] Alan W. Weisgerber,[1] Max D. Palay,[1] Melodie T. Nguyen,[2] Broderick L. Bills,[2] and Michelle K. Knowles[1,2,*]

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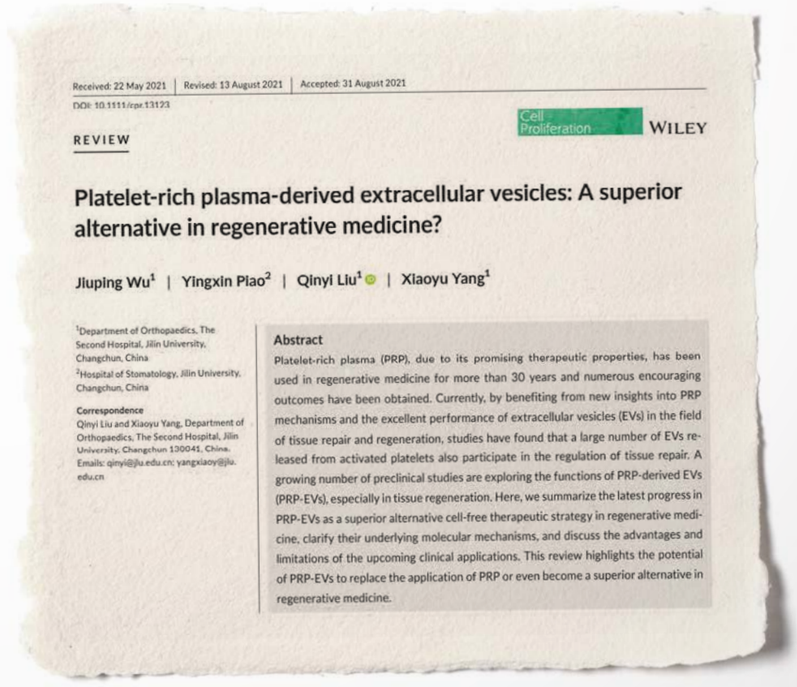
ABSTRACT When multivesicular endosomes (MVEs) fuse with the plasma membrane, exosomes are released into the extracellular space where they can affect other cells. The ability of exosomes to regulate cells nearby or further away depends on whether they remain attached to the secreting cell membrane. The regulation and kinetics of exosome secretion are not well characterized, but probes for directly imaging single MVE fusion events have allowed for visualization of the fusion and release process. In particular, the design of an exosome marker with a pH-sensitive dye in the middle of the tetraspanin protein CD63 has facilitated studies of individual MVE fusion events. Using TIRF microscopy, single fusion events were measured in A549 cells held at 23–37°C and events were identified using an automated detection algorithm. Stable docking precedes fusion almost always and a decrease in temperature was accompanied by decrease in the rate of content loss and in the frequency of fusion events. The loss of CD63-pHluorin fluorescence was measured at fusion sites and fit with a single or double exponential decay, with most events requiring two components and a plateau because the loss of fluorescence was typically incomplete. To interpret the kinetics, fusion events were simulated as a localized release of tethered/untethered exosomes coupled with the membrane diffusion of CD63. The experimentally observed decay required three components in the simulation: 1) free exosomes, 2) CD63 membrane diffusion from the endosomal membrane into the plasma membrane, and 3) tethered exosomes. Modeling with slow diffusion of the tethered exosomes (0.0015–0.004 $\mu m^2/s$) accurately fits the experimental data for all temperatures. However, simulating with immobile tethers or the absence of tethers fails to replicate the data. Our model suggests that exosome release from the fusion site is incomplete due to postfusion, membrane attachment.



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REVIEW

Cell Proliferation WILEY

# Platelet-rich plasma-derived extracellular vesicles: A superior alternative in regenerative medicine?

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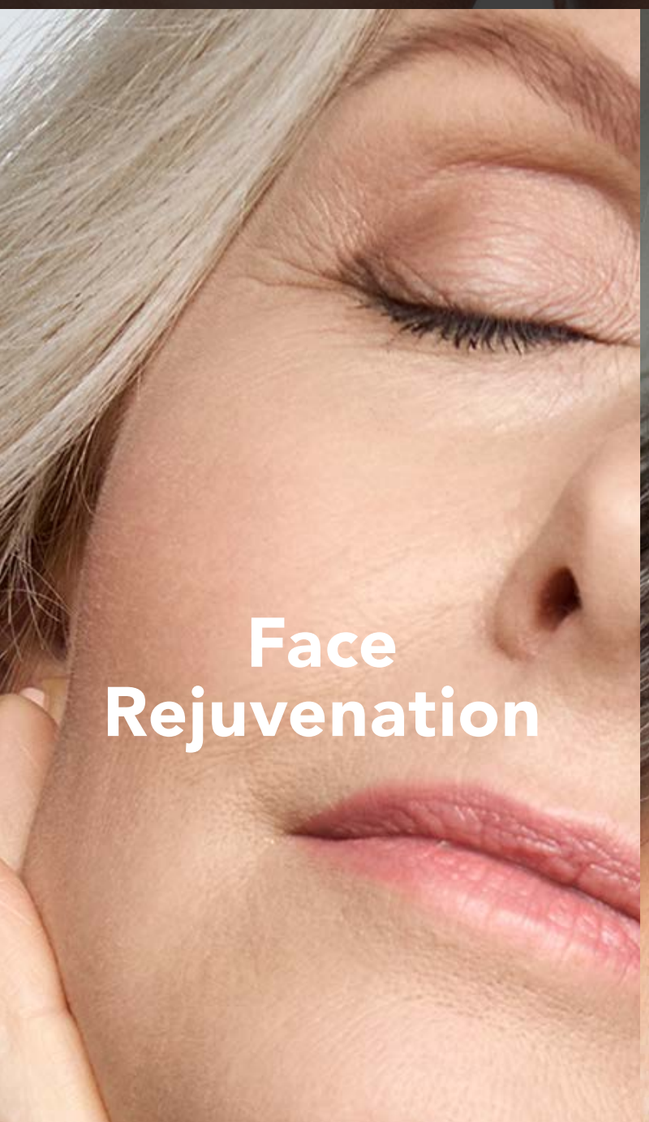
**Abstract**

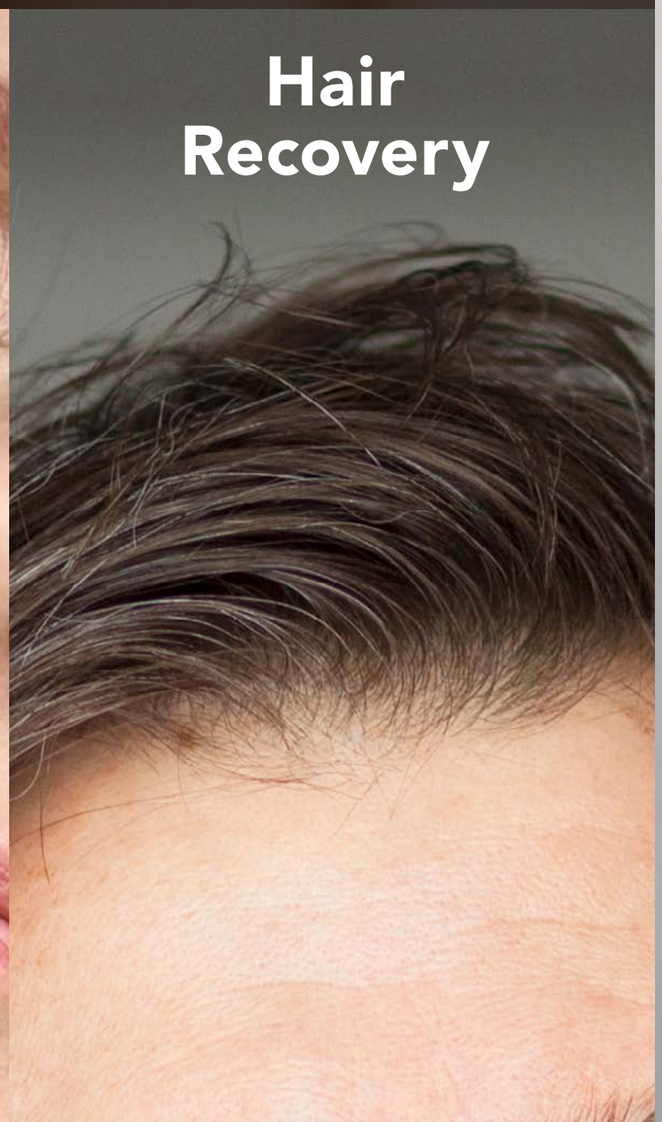
Platelet-rich plasma (PRP), due to its promising therapeutic properties, has been used in regenerative medicine for more than 30 years and numerous encouraging outcomes have been obtained. Currently, by benefiting from new insights into PRP mechanisms and the excellent performance of extracellular vesicles (EVs) in the field of tissue repair and regeneration, studies have found that a large number of EVs released from activated platelets also participate in the regulation of tissue repair. A growing number of preclinical studies are exploring the functions of PRP-derived EVs (PRP-EVs), especially in tissue regeneration. Here, we summarize the latest progress in PRP-EVs as a superior alternative cell-free therapeutic strategy in regenerative medicine, clarify their underlying molecular mechanisms, and discuss the advantages and limitations of the upcoming clinical applications. This review highlights the potential of PRP-EVs to replace the application of PRP or even become a superior alternative in regenerative medicine.

## Highlights

- Blood is one of the richest sources of EVs.
- Cargo and biological properties of EVs are defined by the types and characteristics of parental cells.
- Functions: homeostasis, vascular integrity, inmunoregulation, inflammatory regulation, angiogenesis.
- Cargo: proteins, miRNAs, GFs (bFGF x3.3, TGF-ß1 x35.5).

MCT OBTAINS STUNNING AMOUNTS OF AUTOLOGOUS EXOSOMES FOR SPECTACULAR CLINICAL, ANTIAGING AND AESTHETIC RESULTS




# MCT stand-alone PRESETS

| | Stimulus 1 | Stimulus 2 |
|---|---|---|
| Exosomes | 467 nm<br>2 J/cm²<br>10 min | 37°C<br>10 min |
| PRP | 620 nm<br>1 J/cm²<br>10 min | 4°C<br>15 min |
| Cells | 620 nm<br>1 J/cm²<br>10 min | 850nm<br>0.2 J/cm²<br>10 min |


Leading-edge technology
Evidence-based
Less pain
Safe
Fast
User-friendly

Yang K, Li D, Wang M, Xu Z, Chen X, Liu Q, Sun W, Li J, Gong Y, Liu D, Shao C, Liu Q, Li X. Exposure to blue light stimulates the proangiogenic capability of exosomes derived from human umbilical cord mesenchymal stem cells. Stem Cell Res Ther. 2019.

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# Inspiring technology
# Outstanding results


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