

## Chapter 49

# Can Activated Platelet Rich Plasma Combined with Adipose-Derived Stem Cells Be Used to Treat Skin Wrinkles? A Mechanism Study

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### ABSTRACT

*In recent years, Platelet Rich Plasma (PRP) and Adipose-Derived Stem Cells (ADSCs) have been used separately for many clinical applications, especially skin rejuvenation. A combined injection of PRP and ADSCs could therefore be used to treat skin wrinkles. However, there are controversies and reports with conflicting results regarding the efficacy of this treatment. The authors aimed to determine the anti-wrinkle and skin rejuvenation mechanism of combined PRP and ADSCs treatment. The effects of PRP and ADSCs isolated from the same consenting donors were evaluated using in vitro and in vivo models. The in vitro effects of PRP and ADSCs on dermal fibroblast proliferation, collagen production, and inhibition of Matrix Metalloproteinase-1 (MMP-1) production were investigated using a co-culture model. Fibroblasts and ADSCs were cultured within the same dish, but in two separate cavities (using an insert plate), in the presence of the same PRP-supplemented medium. In vivo, the authors evaluated the effects of combined PRP and ADSCs on skin histochemistry, including changes in the dermal layer and collagen production in photo-aged skin (mice). They also determined the survival and differentiation of grafted ADSCs. The results show that combined PRP and ADSCs strongly stimulate in vitro fibroblast proliferation, collagen production, and inhibition of MMP-1 synthesis. Intra-dermal co-injection of PRP and ADSCs was observed to stimulate increased dermal layer thickness and collagen production compared with the untreated group. These results indicate that a combined PRP and ADSC injection can reduce wrinkles more effectively than either PRP or ADSC alone, and provide insight into the clinical use of PRP combined with ADSCs for dermal applications, particularly skin rejuvenation.*

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## INTRODUCTION

Skin wrinkles are the most obvious sign of ageing skin, usually characterized by small and fine wrinkles, roughness, dryness, laxity, and changes in pigmentation. This phenomenon is due to epidermal thinning and collagen degradation, a process effected and enhanced by ultraviolet rays, so is also referred to as “photo-aged” (Chung, 2001, 2003). Following skin collagen degradation, elasticity is reduced and wrinkles are formed (Fisher, 2008). Wrinkles usually appear on the face skin after the age of 28. The inhibition of skin ageing and therefore wrinkles is of great interest to a majority of people.

Previously, skin wrinkles have been treated using a range of therapies, including collagen, hyaluronic acid, and adipose injections; however, results have been disappointing. Recently, PRP and ADSCs have been used widely across many clinical fields, especially for skincare and cosmetic surgery. PRP is an enriched plasma, obtained by a simple centrifugation technique. It has been reported to stimulate bone adhesion and wound healing (Froum, 2002; Marx, 1998; Petrungaro, 2001; Robiony, 2002; Margolis, 2001; Welsh, 2000; Man, 2001; Bhanot, 2002). PRP contains a pool of at least seven Growth Factors (GFs), including Epidermal Growth Factor (EGF), Platelet Derived Growth Factor (PDGF), Transforming Growth Factor Beta (TGF- $\beta$ ), Vascular Endothelial Growth Factor (VEGF), Fibroblast Growth Factor (FGF), Insulin-like Growth Factor (IGF), and Keratinocyte Growth Factor (KGF). The therapeutic influence of PRP is due to the high concentration of these growth factors compared with normal plasma. Many of these GFs have important roles in wound healing and tissue regeneration. EGF can stimulate proliferation, differentiation, and survival of cells (Herbst, 2004). KGF, also known as FGF7, is a GF present in the epithelialization phase of wound healing. It stimulates keratinocyte proliferation to cover the wound and form the epithelium. These GFs are reported to improve

photo-aged skin, enhance collagen production, and stimulate keratinocyte proliferation within skin (Fitzpatrick, 2003; Ehrlich, 2006; Gold, 2007). Results from previous studies have shown PRP to have many benefits for skin regeneration and anti-aging. PRP injection into nude mice resulted in rejuvenate photo-aged skin (Cho, 2011). PRP has also been shown to stimulate expression of type I collagen (col-I) and MMP-1 protein in dermal fibroblasts (Kim, 2011) and to increase expression of G1 cycle regulators, col-I, and MMP-1 to accelerate wound healing (Cho, 2012). PRP used in combination with fractional laser treatment has been successful for skin rejuvenation. This combination can stimulate keratinocyte and fibroblast proliferation and collagen production, so increase dermal elasticity.

Recently, Adipose-Derived Stem Cells (ADSCs) were used for skin rejuvenation and anti-aging applications. ADSCs are multipotent stem cells derived from adipose tissue (Zuk, 2002; Bunnell, 2008; Witkowska-Zimny, 2011; Yu, 2011). ADSCs can differentiate into various lineages, including adipocytes (Chen, 2012); insulin producing cells (Kim, 2010), endothelial cells (Colazzo, 2010; Colazzo, 2010; Zhang, 2011; Marino, 2012); osteoblasts (Xu, 2005; Safwani, 2011), hepatocytes (Al Battah, 2011; Snykers, 2011); chondrocytes (Estes, 2010; Boeuf, 2010; Musumeci, 2011), and neuronal-like cells (Gardin, 2011). ADSC transplantation has been reported to benefit skin rejuvenation by triggering dermal wound healing and inhibiting photo-aging (Kim, 2007) and also by inhibiting UVB-induced wrinkles and stimulating collagen synthesis (Kim, 2009, 2011). Furthermore, ADSCs are reported to improve autologous fat transplantation, exhibit an antioxidant action and whitening effect (Matsumoto, 2006; Kim, 2008) and increase angiogenesis (Kim, 2011). There exist two reported mechanisms explaining how ADSCs provide these therapeutic effects on skin. The first mechanism relates to secretory factors produced by ADSCs, including Hepatocyte Growth Factor (HGF),

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