The Effect of Platelet Rich Plasma on the Treatment Outcome of Minigraft/NB-UVB Therapy in Stable Vitiligo: Clinical Evaluation and Effect on Tissue levels of bFGF

Thesis

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Tabarek Abdulkhaleq

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List of Abbreviations

Abb.	Full term
6-BH4	6R-L-erythro-tetrahydrobiopterin
APS	Autoimmune polyglandular syndrome
bFGF	Basic fibroblast growth factor
CDK4	Cyclic dependent kinase 4
CES	Cultured epithelial suspension
CIE	Commission international de l'eclairage
CMS	Cultured melanocyte suspension
DS	Double spin
EGF	Epidermal growth factor
FAK	Antifocal adhesion kinase
FGF	Fibroblast growth factor
FGFR	Fibroblast growth factor receptor
FOXP3	Forkhead box P3 master regulator of the Treg cell
G1 phase	Gap 1 phase
GF	Growth factor
GMC-SF	Granulocyte Monocyte Colony Stimulating Factor
gp100	Glycoprotein 100
H_2O_2	Hydrogen peroxide
HGF	Hepatocyte Growth Factor
HLA-A2	Human leukocyte antigen-A2
HS	Highly significant
ICAM-1	Intracellular adhesion molecule-1
IFN-γ	Interferon gamma
IGF	Insulin like growth factor
IL	Interleukin
IQR	Interquartile range
JNK	C. Jun NH2-terminal kinase protein

List of Abbreviations Cont...

Abb.	Full term
KGF	Keratinocyte growth factor
MART-1	Melanoma antigen recognized by T cells 1
MBEH	Monobenzyl Ether of hydroquinone
MC1R	Melanocortin 1 receptor
MED	Minimal erythemal dose
MG	Minigraft
NCES	Non-cultured epidermal suspension
NO	Nitric oxide
NS	Non significant
NSV	Non-segmental vitiligo
O.D	Optic density
ONOO-	Peroxynitrite anion
PASI	Psoriasis area severity index
PBS	Phosphate buffered saline
PDGF	Platelet-derived growth factor
PI3K/AKT	Phosphatidylinositol-3-kinase/serine/threonine protein kinase
PPP	Platelet poor plasma
pRb	Retinoblastoma protein
PRI	Potential regimentation index
PRP	Platelet rich plasma
PSG	Partial thickness skin graft
PUVA	Psoralen plus ultraviolet A
r.p.m	Revaluation per minute
Rac1	Guanosine triphosphatases (GTpases) protein
RNS	Reactive nitrogen species
ROS	Reactive oxygen species
RPE	Retinal pigment epithelium

List of Abbreviations Cont...

Abb.	Full term
r_s	Spearman's correlation coefficient
S	Significant
S. phase	DNA synthsis phase
SBEG	Suction blister epidermal graft
SCF	Stem cell factor
SD	Standard deviation
SS	Single spin
STSG	Split thickness skin graft
SV	Segmental vitiligo
TGF-β	Transforming growth factor beta
TNF-α	Tumor necrosis factor-α
TPA	Tetradecanoylphorbol acetate
Tregs	Regulatory T
TRP-1	Tyrosinase-related protein one
TRP-2	Tyrosinase-related protein two
U-test	Mann-whitney test
UV	Ultraviolet
UVA	Ultraviolet-A
UVB	Ultraviolet B
VASI	Vitiligo area scoring index
VEGF	Vascular endothelial growth factor
VESTA	Vitiligo extent score for a target area
VETF	Vitiligo European taskforce assessment
VETI	Vitiligo extent-tensity index
VIDA	Vitiligo disease activity score

1. INTRODUCTION

Vitiligo is an acquired pigmentary disorder resulting from loss of melanocytes which causes depigmentation of the skin. Clinically, it is characterized by the progressive loss of melanocytes causing the appearance of well-circumscribed milky white cutaneous macules and patches ***(Felsten et al., 2011).*** The worldwide prevalence is estimated to be 0.5-2% of the general population ***(Krüger and Schallreuter, 2012)***. Vitiligo is generally classified in three forms: non-segmental vitiligo (NSV), a group which includes the acrofacial, mucosal, generalized, universal and mixed forms; segmental vitiligo (SV), which can be unisegmental, bisegmental, or multisegmental; and the unclassified or undetermined form ***(Faria et al., 2014)***.

The exact etiology of vitiligo has been proposed of many competing theories. The autoimmune theory is the most accepted one, being sustained by several epidemiological, clinical, and experimental findings ***(Patel et al., 2017)***. It is suggested that melanocyte destruction in susceptible individuals is triggered through an autoimmune response. Several exogenous and endogenous stimuli have been linked to the onset of the disease. The exogenous factors include ultraviolet irradiations, trauma (Koebner phenomenon), stress, major infections, malignancies, neural abnormalities, calcium imbalance, certain drugs, hormones, and exposure to cytotoxic

compounds ***(Rodrigues et al., 2017).*** All are thought to induce oxidative stress in melanocytes, as indicated by the high levels of reactive oxygen species (ROS), mainly hydrogen peroxide, found in lesional skin ***(Sravani et al., 2009; Picardo and Bastonini, 2015).***

Treatment for vitiligo is difficult and often challenging. Treatment options include corticosteroids, calcineurin inhibitors, vitamin D analogues, antioxidants, phototherapy using ultraviolet B (UVB), psoralen plus ultraviolet A (PUVA), narrow-band UVB, excimer laser ***(Felsten et al., 2011; Patel et al., 2012)***.

Surgical alternatives represent good options for patients not responding to the above-mentioned therapies; however, surgery is limited to segmental or localized vitiligo. The main purpose of the surgical treatment is to restore melanocytes on the target sites. The repigmentation process should begin when the transplanted cells survive the procedure since melanin is transferred by melanocytes to keratinocytes pass through the dendritic process, giving the skin color and natural appearance. There are 5 basic methods for repigmentation surgery which are thin dermoepidermal grafts, suction epidermal grafting, punch minigraft, noncultured epidermal suspension and cultured melanocyte suspension ***(Van Geel et al., 2001)***.

Among these surgical approaches, punch minigrafting represents a simple and effective procedure, which involves performing multiple perforations in the recipient

depigmented site using biopsy punches and transplanting similar size punches taken from a normally pigmented donor site. Punch grafting followed by NB-UVB was found to be effective in inducing repigmentation in vitiligo patients ***(Lahiri et al., 2006).***

Platelet-rich plasma (PRP) represents a bio technology that is part of the growing interest in tissue engineering and cellular therapy. PRP is an autologous preparation of platelets in concentrated plasma. Although the optimal PRP platelet concentration is unclear, the current method by which PRP is prepared is reported to involve 300-700% enrichment, with high platelet concentrations ***(Sclafani, 2009)***. PRP has emerged as an attractive treatment modality for many dermatological conditions like treatment acne scars, striae distensae, wound healing, hair re-growth ***(Casabona et al., 2010).*** The beneficial effects of PRP are thought to be attributed to the released growth factors from the activated platelets ***(Hara and Basu, 2014).*** Various growth factors have been described to be released from the activated platelets including platelet-derived growth factor, transforming growth factor, vascular endothelial growth factor, basic fibroblast growth factor, insulin like growth factor, epidermal growth factor and connective tissue growth factor ***(Marx, 2004).*** These factors are known to regulate many processes including cell migration, attachment, proliferation, differentiation, and promoting extra cellular matrix accumulation by binding to specific cell-surface receptors ***(Wrotniak et al., 2007).***