Extracorporeal Photopheresis Past, Present and future

ESSAY

Submitted for partial fulfillment of the Master degree in Dermatology, Venereology and Andrology

Presented By

Norhan Nagi Saad El Deen Hamed

M.B., B.Ch. Faculty of Medicine, Ain Shams University

Supervised By

Prof. Dr. Mohamed Abd El Naeem Sallam

Professor of Dermatology, Venereology & Andrology Faculty of Medicine, Ain Shams University

Dr. Khaled Mohamed Abd El Raouf

El Zawahry

Lecturer of Dermatology, Venereology & Andrology Faculty of Medicine, Ain Shams University

> Ain Shams University-2012

فصل مكونات الدمر خارج الجسم وتعريضها للأشعة الضوئية في الماضي والحاضر والمستقبل

رسالة مقدمة كجزء متمم للحصول على درجة الماجستير في الأمراض الجلدية والتناسلية وأمراض الذكورة

> مقدم من الطبيبة **نور هان ناجي سعد الدين** بكالوريوس الطب والجراحة – جامعة عين شمس

تحت إشراف **الأستاذ الدكتور/ محمد عبد النعيم سلام** أستاذ الأمراض الجلدية والتناسلية وأمراض الذكورة كلية الطب – جامعة عين شمس

الدكتور/ خالد محـمد عبد الرؤوف الظواهري

مدرس الأمراض الجلدية والتناسلية وأمراض الذكورة كلية الطب – جامعة عين شمس

Summary

Extracorporeal photopheresis (ECP) is defined as an immuno-modulatory technique that involves removal of peripheral blood, separation of the buffy coat. and photoactivation with a photosensitizer (8-MOP) and ultraviolet A irradiation before re-infusion of cells. It was developed more than 20 years ago by Edleson to treat erythrodermic T cell lymphoma (CTCL).

Most of studies revealed that the mechanism of action of ECP is to induce leucocyte apoptosis, followed by their engulfment by macrophages or other antigen presenting cells, such as immature dendiritic cells in an anti inflammatory cytokin environment. The anti inflammatory cytokine secretion pattern, with a switch from TH1to TH2 for CD4+ lymphocytes, and the engulfment by immature cells without co-stimulatory molecules induces anergy, by deleting effector T- cells that responded to the presented antigens. An increase in regulatory T- cells is also induced after ECP and may contribute to allograft acceptance by the recipient.

However other studies suggest that mechanism of ECP also involves the recruitment and involvement of additional immune cells. Till now it is clear ECP, both activates tumor immunity

Acknowledgment

First of all, I would like to express my endless and everlasting thanks to **MIGHTY GOD ALLAH**; without his help, this work would never have been finished.

Although no words can express my great gratitude and respect to **Prof. Dr. Mohamed Abd El naeem Sallam**; Professor of Dermatology, Venereology & Andrology, Ain Shams University, I would like to thank him for his outstanding encouragement, advice and his sincere endless support throughout this work.

I feel greatly indebted to **Dr. Khaled Mohamed Abd El Raouf El Zawahry**; Lecturer of Dermatology, Venereology & Andrology, Ain Shams University, for his great care, patience, sincere guidance, tremendous effort and continuous valuable advice throughout this work.

Norhan Nagi Saad El Deen Hamed

CONTENTS

Title	No
List of Abbreviations	i
List of Figures	v
List of Tables	vii
Introduction	1
Aim of the Work	4
Review of literature	
Chapter One:	5
• Definition of ECP	5
History of ECP	8
Chapter Two: Mechanism of Action of ECP	10
Chapter Three: Indications of ECP	35
Chapter Four: ECP techniques & methods	80
Chapter Five: Safety and Side effects of ECP	91
Chapter Six: ECP in children and low body weight patients	96
Summary	100
References	103
Arabic Summary	

List of abbreviations

Abbrev	Meaning
8-MOP	8-methoxypsoralen
aGVHD	acute graft versus host disease
AMR	Antibody mediated rejection
Apaf-1	Apoptotic protease activating factor 1
APCs	Antigen presenting cells
Apo2l	Apoptosis ligand 2
Bad	Bcl ₂ – associated death prompter
Bak	Bcl2 – homologous antagonist killer
Bax	Bcl_2 – associated x protein
BCL- XL	B cell leukemia X long
BCL	B cell leukemia 2
BH3	BCL homology
Bid	BH3 domain only death against protein
Bim	Bcl ₂ interacting mediator
BIRs	baculoviral repeats
Bmf	Bcl ₂ modifying factor
BRUCE	BIR repeat containing ubiquitin conjugating
	Enzyme system
BOS	Bronchiolitis obliterans syndrome
Caspases	cysteine aspartic acid Specific proteases
CD4	Cluster of differentiation
CELLEXTM	cell extensible access method
c-FLIP	c-FLICE inhibitory protein

cGVHD	chronic graft versus host disease
СНОР	Cyclo-phosphamide doxorubicin vincristine
	prednisone
cIAP	Cellular IAP
CMV	Cytomegalo virus
CR	Complete response
CTA	Composite tissue allotransplantation
CTCL	Cutaneous T cell lymphoma
DCs	Dendiritic cells
DDCs	Dermal dendiritic cells
DISC	Death inducing signaling complex
DNA	Deoxyribonucleic acid.
DR	Death receptor
ECP	Extracorporeal photopheresis
ECV	Extracorporeal volume
FADD	Fas-associated death domain
Fas L	Fas ligand
FDA	Food and Drug Administration.
FoxP3	Fork head box P3
GAS6	Growth arrest specific 6
GMP	Good manufacturing practice
HCV	Hepatitis C virus
HLA	Human leukocyte antigen
HMGB1	High mobility group box 1
HSP	Heat shock protein
HSV1	Herpes simplex virus type1

IAPs	Inhibitors of apoptosis proteins
IgE	Immunoglobulin E
ILP	IAP like protein
IL10	Interleukin 10
INF	Interferon
IRF4	Interferon regulatory factor 4
LCs	Langerhans cells
mDCs	Myeloid dendiritic cells
MF	Mycosis fungoides
MFG	Mobile fat globulin
MLIAP	Melanoma IAP
MLR	Mixed lymphocyte reaction
NFĸB	Nuclear factor kappa B.
NIH	National institutes of health
NOXA	Naphthoxy acetic acid
PAF	Platelet activating factor
PBMC	peripheral blood mononuclear cells
PCD	programmed cell death
pDCs	plasmacytoid dendiritic cells
PF	Pemphigus foliaceus
PI	Propidium iodine
Pre-PCD	Pre programmed cell death
PRP	Pityriasis rubra pilaris
PS	Phosphatidylserine
PUMA	P ₅₃ Upregulated modulator of apoptosis
PUVA	Psoralen ultraviolet A irradiation

PV	Pemphigus vulgaris
RBCs	Red blood cells
RCTs	Randomized controlled trials
SLE	Systemic lupus erythematosus
SMAC	Second mitochondria derived activator of
	caspases.
SS	Se'zary syndrome
Sulfo Lac Nac (slav	ı) sulfo N acetyllactosamine
TAM	Tyr 3, axl, mer
TBV	Total blood volume
TGF-B1	Transforming growth factor B1
Th	T-helper
TIM4	T cell immunoglobulin mucin 4
TLR	Toll-like receptor
TNF	Tumor necrosis factor
TPE	Therapeutic plasma exchange
TRAIL	Tumor necrosis factor related apoptosis
	inducing ligand
UVA	Ultraviolet A
UVADEX	Brand name of methoxsalen
UVAR –XTS	Ultra violet Auto blood Radiation xytex
	tissue services.
UVAR	Ultraviolet A radiation
UVB	Ultraviolet B
WBCs	White blood cells
XIAP	X-linked inhibitor of apoptosis protein

List	of	Figures	5
------	----	---------	---

Fig. No.	Title	Page
Figure 1:	Diagram to show ECP technique	7
Figure 2:	Principles of extracorporeal photopheresis in	11
	composite tissue allotransplantations	
Figure 3:	Elements of the extrinsic apoptotic pathway	13
Figure 4:	FADD recruitment and caspase activation via the	16
	extrinsic pathway	
Figure 5:	Elements of the intrinsic apoptotic pathway.	17
Figure 6:	Elements assemble to form the apoptosome.	20
Figure 7:	The caspase cascade.	21
Figure 8:	IAPs and caspase inhibition.	24
Figure 9:	Time course of the release of blebs of	25
	lymphocytes after treatment with 8-MOP or UVA	
	A : In non stimulated cells	
	B : In stimulated cells	
Figure 10:	Modulation of the cytokine profile and the T	33
	lymphocyte compartment by tolerogenic dendritic	
	cells generated by photopheresis	

v

- Figure 11: clinical presentation of late stage nephrogenic 49 systemic fibrosis
 A: significant skin sclerosis affecting the lower extremities
 B: sever skin contractures of the ankle joints
- Figure 12: Kaplan-Meier probability of overall survival 57 among patients with steroid-refractory and dependent acute graft-versus-host disease
- Figure 13: Cumulative incidence of complete or partial 60 response of skin was higher among patients with steroid refractory/- dependent/-intolerant chronic graft-versus host disease treated with extracorporeal photopheresis (ECP) than in the control group

Figure 14:	UVAR tech	nique first	generation		82
Figure 15:	UVAR tech	nique seco	ond generation		85
Figure 16:	off-line tech	nique			87
Figure 17:	therapeutic	cellular	immunomodulation	using	88

extracorporeal photochemotherapy

 Table 1: Pro- and anti-apoptotic members of the Bcl-2 superfamily of proteins. Table 2: Diseases treated with extracorporeal photopheresis Table 3: ECP is a recommended therapy for the first line treatment of mycosis fungoid stage III and sezary syndrome Table 4: Comparison between standard therapy alone and standard therapy with ECP Table 5: implementation of ECP in solid organ transplantation for the treatment of acute rejection episodes 	Table No.	Title	Page
 Table 2: Diseases treated with extracorporeal photopheresis Table 3: ECP is a recommended therapy for the first line treatment of mycosis fungoid stage III and sezary syndrome Table 4: Comparison between standard therapy alone and standard therapy with ECP Table 5: implementation of ECP in solid organ transplantation for the treatment of acute rejection episodes 	Table 1:	Pro- and anti-apoptotic members of the Bcl-2	18
 Table 2: Diseases treated with extracorporeal photopheresis Table 3: ECP is a recommended therapy for the first line treatment of mycosis fungoid stage III and sezary syndrome Table 4: Comparison between standard therapy alone and standard therapy with ECP Table 5: implementation of ECP in solid organ transplantation for the treatment of acute rejection episodes 		superfamily of proteins.	
 Table 3: ECP is a recommended therapy for the first line treatment of mycosis fungoid stage III and sezary syndrome Table 4: Comparison between standard therapy alone and standard therapy with ECP Table 5: implementation of ECP in solid organ transplantation for the treatment of acute rejection episodes 	Fable 2:	Diseases treated with extracorporeal photopheresis	36
 treatment of mycosis fungoid stage III and sezary syndrome Table 4: Comparison between standard therapy alone and standard therapy with ECP Table 5: implementation of ECP in solid organ transplantation for the treatment of acute rejection episodes 	Fable 3:	ECP is a recommended therapy for the first line	42
 syndrome Table 4: Comparison between standard therapy alone and standard therapy with ECP Table 5: implementation of ECP in solid organ transplantation for the treatment of acute rejection episodes 		treatment of mycosis fungoid stage III and sezary	
Table 4:Comparison between standard therapy alone and standard therapy with ECPTable 5:implementation of ECP in solid organ transplantation for the treatment of acute rejection episodesTable 6:Side off the state is the ide ECP		syndrome	
standard therapy with ECP Table 5: implementation of ECP in solid organ transplantation for the treatment of acute rejection episodes Table 6: Side off the treatment of acute rejection the treatment of acute rejection episodes	Fable 4:	Comparison between standard therapy alone and	65
Table 5: implementation of ECP in solid organ transplantation for the treatment of acute rejection episodes Table 6 Side off the treatment of acute rejection		standard therapy with ECP	
transplantation for the treatment of acute rejection episodes	Fable 5:	implementation of ECP in solid organ	68
episodes		transplantation for the treatment of acute rejection	
		episodes	
Table 6: Side effect associated with ECP	Fable 6:	Side effect associated with ECP	95

List of Tables

Introduction

Extracorporeal photopheresis (ECP) also known as extracorporeal photochemotherapy, it is an immunomodulatory technique based on pheresis of light sensitive cells. It is consists of exposure of peripheral blood mononuclear cells (PBMC), collected by aphaeresis, to ultraviolet A light in the presence of the DNA-intercalating agent 8-methoxypsoralen. The treated cells undergo apoptosis and are readily reinjected into the patient, leading to antigen-specific immunomudulation (*Aubin and Mousson, 2004*). This technique is reported by Edleson et al in 1987 to treat erythrodermic cutaneous T cell lymphoma (*Edleson et al., 1987*)

However it has been reported later on to be effective for a wide variety of disease such as acute graft versus host disease (GVHD), solid organ transplant rejection, crohn's disease, scleroderma, diabetes mellitus, multiple sclerosis, systemic sclerosis, bullous pemphigoid, pemphigus vulgaris, pityriasis rubra pilari, nephrogenic systemic fibrosis. Pemphigus foliaceus, systemic lupus erythromatosis, psoriatic arthritis, psoriasis vulgaris, rheumatoid arthritis, atopic dermatitis, juvenile dermatomyositis, scleromyxedema, and most widely in steroid refractory chronic graft versus host disease (cGVHD).(*Maeda, 2009; Chiesa-Fuxench and Gonzalez Chavez, 2010*)

Further more, the use of ECP in some of these conditions may allow a significant reduction in the use of systemic steroid and other immunosuppressant, reducing long term morbidity, and mortality (*Knobler et al., 2009*)

The advantage of the photopheresis treatment is the low frequency of side effects such as vasovagal syncope or infection, the disadvantages however are the practical efforts required and the high treatment cost (*Meada, 2009*)

Extracorporeal photopheresis is performed using the UVAR XTS Photopheresis System developed by Therakos, the process is performed through one intravenous access port and has 3 basic stages: (1) leukapheresis, (2) photoactivation, and (3) reinfusion. The process takes 3-4 hours to complete.

- One 16-gauge peripheral intravenous line or central venous access is established in the patient.
- Blood (225 mL) is passed through 3 cycles of leukapheresis, or 125 mL of blood is passed through 6 cycles, depending on the patient's hematocrit value and body size. At the end of each leukapheresis cycle, the red blood cells and plasma are returned to the patient.
- The collected WBCs (including approximately 5% of the peripheral blood mononuclear cells) are mixed with heparin, saline, and 8-methoxypsoralen (8-MOP), which intercalates into the DNA of the lymphocytes upon

exposure to UVA light and makes them more susceptible to apoptosis when exposed to UVA radiation.

- The mixture is passed as a 1-mm film through a sterile cassette surrounded by UVA bulbs for 180 minutes, resulting in an average UVA exposure of 2 J/cm² per lymphocyte.
- The treated WBC mixture is returned to the patient. (*Camile et al., 2011*)

It has been suggested that ECP therapy, unlike other regimens, immunosuppressive does not cause global immunosuppression, but induces immune tolerance. Recent clinical and animal studies demonstrate that ECP therapy induces antigen-specific regulatory T cells, including CD4, CD25, FoxP3, T cells and IL-10-producing Tr1 cells, that may arise secondarily to the induction of tolerogenic antigenpresenting cells (APCs) by infusion of apoptotic cells. It has also been suggested that ECP therapy may induce IL-10producing regulatory B cells and regulatory CD8 T cells. Finally, several recent studies, which examined the cellular elements involved in the uptake of apoptotic cells, demonstrated that apoptotic cells modulate APCs through binding to specific receptors, particularly TAM receptors that provide inhibitory signals that block APC activation. (Chang-*Qing Xia et al.*, 2009)