The Bcl 1 Single Nucleotide Polymorphism of the Human Glucocorticoid Receptor Gene h-GR/NR3C1 Promoter in Patients with Bronchial Asthma

Thesis

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This work is dedicated to . . .

My beloved father & uncles, ***for always being there for me***

My mother ***to whom I owe everything I ever did in my life and will achieve***

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List of Abbreviations

Abb.	Meaning

AC……: Adenylyl cyclase.
ACTH ..... : Corticotrophin.
AF1 ..... : Activation function domain.
AHR..... : Airway hyperresponsiveness.
ANA ..... : Antinuclear antibodies.
AP-1..... : Activator protein 1.
APCs..... : Antigen presenting cells.
APOE ..... : Apolipoprotein E.
ARMS..... : Amplification Refractory Mutation System.
ASM..... : Airway smooth muscle.
ASO ..... : Allele specific oligonucleotide.
ATP..... : Adenosine triphosphate.
BA..... : Bronchial asthma.
BAL ..... : Bronchoalveolar lavage.
BMI..... : Branch Migration Inhibition.
cAMP..... : Cyclic adenosine monophosphate.
CBG..... : Corticosteroid binding globulin.
CCD..... : Cross-reactive carbohydrate determinants.
cGCR..... : Cytosolic glucocorticoid receptor.
CLA ..... : Chemiluminescent assay.
CNS ..... : Central nervous system.
COPD ..... : Chronic obstructive pulmonary disease.
COX..... : Cyclooxygenase.
CRP ..... : C - reactive protein.
CT..... : Computed Tomography**.**
CysLTs..... : Cysteinyl leukotriences.
DBD..... : DNA-binding domain.
DCs..... : Dendritic cells.
ECM..... : Extracellular matrix.
EIB ..... : Exercise-induced bronchoconstriction.
ELISA..... : The enzyme-linked immunosorbent assay.

List of Abbreviations (Cont...)

Abb.	Meaning

ENFUMOSA ..... : European Network for Understanding Mechanisms of Severe Asthma.
eNOS ..... : Endothelial nitric oxide synthase.
ERK ..... : Extracellular signal-regulated kinases.
FcεRI ..... : Fc epsilon receptor I.
FEV ..... : Forced Expiratory Volume.
FRET ..... : Fluorescence resonance energy transfer.
FVC ..... : Forced Vital Capacity.
GC ..... : Glucocorticoid.
GCR ..... : Glucocorticoid receptor.
GINA ..... : The Global Initiative for Asthma.
GM-CSF ..... : Granulocyte-macrophage colony-stimulating factor.
GREs ..... : Glucocorticoid Response Elements.
Gs protein ..... : G stimulatory protein.
HDAC2 ..... : Histone deacetylase.
HPA ..... : Hypothalamic pituitary adrenal axis.
HRCT ..... : High-resolution computed tomography.
Hsp90 ..... : Heat shock protein 90.
ICAM ..... : Intercellular cell adhesion molecule.
IGF ..... : Insulin-like growth factor.
IGF-dependent IGFBP-4 Protease: Insulin-like growth factor-dependent-binding protein-4 protease.
IL ..... : Interleukin.
INF ..... : Interferon.
JAK ..... : Janus-kinase.
JNKs ..... : Jun amino-terminal kinases.
LABA ..... : Long-acting beta-adrenoceptor agonists.
LBD ..... : Ligand binding domain.
LPS ..... : Lipopolysaccharides.
MAPK ..... : Mitogen activated protein kinase.
MAST ..... : The Multiple Allergen Simultaneous Test.

List of Abbreviations (Cont...)

Abb.	Meaning

MBP..... : Major basic protein.
mCD14 ..... : Membrane-bound CD14.
MCP..... : Monocyte chemotactic protein.
mDCs..... : Myeloid dendritic cells.
MDR ..... : Multidrug resistance gene.
MIP..... : Macrophage inflammatory protein.
MMP-9 ..... : Matrix metalloproteinase-9
mRNA........: Messenger RNA.
NF-□□.....: Nuclear factor kappa beta.
nGRE ..... : Negative GRE.
NK..... : Natural killer.
NLSs..... : Nuclear localization signals.
NR3C1..... : Nuclear receptor subfamily 3, group C, member 1
NOS ..... : Nitrous Oxide Systems.
OLA ..... : Oligonucleotide ligation assay.
PAF..... : Platelet activating factor.
PAPPA..... : Pregnancy-associated plasma protein A.
pDCs..... : Plasmacytoid dendritic cells.
PEF..... : Peak expiratory flow.
PGD2..... : Prostaglandin D2.
PKA ..... : Protein kinase A.
PRRs ..... : Pattern recognition receptors.
QTL ..... : Quantitative trait loci.
RANTES ..... : Regulated on activation normal T expressed.
RAST..... : Radioallergosorbent test.
RFLP..... : Restriction fragment length polymohrpisms.
SABA..... : Short acting $beta_2$-adrenoceptor agonists.
SBM..... : Subepithelial basement membrane.
sCD14..... : Soluble CD14.
SNP ..... : Single nucleotide polymorphism.
SRA..... : Steroid resistant asthma.

List of Abbreviations (Cont...)

Abb.	Meaning

SSCP ..... : Strand Conformation Polymorphism.
STAT ..... : Signal transducers and activators of transcription.
TAE……: Tris -Acetate- EDTA.
TCA ..... : Tricyclic antidepressants.
TGF ..... : Transforming growth factor.
TIMP-1..... : Tissue inhibitor of metalloproteinases-1.
TLRs..... : Toll-like receptors.
TNF ..... : Tumor necrosis factor.
TSLP …… : Thymic stromal lymphopoietin.
Tyk ..... : Tyrosine kinase.
VCAM..... : Vascular cell adhesion molecule.
ZO ..... : Zona occludens.

1 INTRODUCTION

Bronchial asthma is a disease with multifactor etiology. The hereditary component of asthma is determined polygenetically. The environmental component is implied significantly by neuroimmune reactions occurring at the molecular level. It should be emphasized that bronchial asthma is a disorder whose primary cause can probably be traced in the disturbed immunoregulatory mechanisms at the lymphocyte level, with secondary overproduction of IgE class antibodies and allergic inflammatory condition ***(Pietras et al., 2011).***

The clinical presentation of bronchial asthma is a resultant of interactions between environmental and genetic factors ***(Panek et al., 2012).*** Multifactor character of etiopathogenesis of bronchial asthma results in significant hereditary variability of phenotypic traits ***(Panek et al., 2012).*** The role of genetic factors in etiopathogenesis of bronchial asthma is currently estimated to range from 36 to 94% ***(Anderson, 2008).***

Glucocorticosteroids (GCs) constitute the basic group of medications used to control inflammatory conditions in patients with bronchial asthma. They exert a multidirectional and specific effect on various cell types. They regulate the expression of specific genes within the cell nuclei via the corticosteroid/ corticosteroid receptor complex (GCs/GCR) ***(Pietras et al., 2011).***

Glucocorticosteroid resistance is a complex problem. It may be constitutional in character, or develop as a sequel to an inflammatory process. It should be emphasized that each tissue represents different sensitivity to GCs. There are serious doubts whether we are born with steroid resistant asthma, or we acquire it during our lifetime ***(Pietras et al., 2011).***

The human glucocorticoid receptor gene also known as nuclear receptor subfamily 3, group C, member 1 gene (NR3C1) is the receptor to which cortisol and other glucocorticoids bind and located on chromosome 5q31-q32 and consists of 9 exons ***(Hawkins et al., 2004).***

The molecular mechanism of action of glucocorticoids involves binding of the specific ligand/glucocorticoid receptor to the sequence of regulator genes encoding the synthesis of the anti-inflammatory proteins determining the clinical effects of glucocorticoids ***(Pietras et al., 2011).***

Polymorphisms present within the h-GR/NR3C1 gene may inhibit formation of GCR/GCs complexes reduce transcription and cause transrepression of the genes encoding proteins synthesized within framework of cellular response to GCs ***(Maltese et al., 2009).***

Bcl1 restriction fragment length polymorphism (RFLP) is formed as a result of changes in a single base. A single nucleotide polymorphism (SNP) involves substitution of a nucleotide by

another one within the h-GCR/NR3C1 gene changes the sense of nucleotide sequence and regulatory regions of the gene modifying its expression. Structural changes within the NR3C1 gene lead to the formation of various alleles which may be associated with resistance to glucocorticoids and development of severe/difficult form to treat asthma ***(Panek et al., 2012).***