

**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

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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 leukotrienes.
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-κ	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 polymorphisms.
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.

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*).