Prognostic Impact of Isocitrate Dehydrogenase Enzyme Isoforms IDH1 & IDH2 Mutations in Acute Myeloid Leukemia

Thesis

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بسم الله الرحمن الرحيم
وَأَنْزَلَ اللهُ عَلَيْكَ
الْكِتَابَ وَالْحِكْمَةَ
وَعَلَّمَكَ مَا لَمْ
تَكُنْ تَعْلَمُ وَكَانَ
فَضْلُ اللهِ عَلَيْكَ
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List of Abbreviations

Abb.	Full term
2-HG	*2-hydroxyglutarate*
5mC	*5-methylcytosine*
ADH	*Antidiuretic hormone*
ALC	*Absolute lymphocyte count*
AML	*Acute myeloid leukemia*
AML-MRC	*Myelodysplasia-related changes*
APL	*Acute promyelocytic leukemia*
ASXL1	*Additional Sex Comb-Like 1*
ATRA	*All trans-retinoic acid*
AUC	*Area under the ROC curve*
Ca	*Calcium*
CBC	*Complete blood picture*
CBFA2	*Core-binding factor subunit α-2*
CD	*Common differentiation*
CEBPA	*CCAAT Enhancer Binding Protein α*
CEBPA	*CCAAT enhancer-binding protein α*
CIBMTR	*Center for international blood and marrow transplant research*
CN-AML	*Cytogenetically normal AML patients*
CR	*Complete remission*
CRi	*Complete remission with incomplete hematologic recovery*
CyTOF	*Cytometry by time-of-flight*
DFN	*Difference from normal*
DIC	*Disseminated intravascular coagulopathy*
DNA	*Deoxyribonucleic acid*
DNMT3A	*DNA-Methyltransferase 3A gene*
ELN	*European Leukemia Net*
EM	*Electron microscopy*
FISH	*Fluorescence in situ hybridization*
FITC	*Fluorescein isothiocyanate*
FLT3	*Fms-Like Tyrosine Kinase 3*

List of Abbreviations cont...

Abb.	Full term
FLT3/ITD	*FMS-like tyrosine kinase 3/ Internal tandem duplications*
FN	*False negative*
FP	*False positive*
HCT	*Hematopoietic cell transplantation*
HLA-DR	*Human leukocyte antigen-antigen D related*
HMAs	*Hypomethylating agents*
HSPCs	*Hematopoietic stem/progenitor cells*
IDH	*Isocitrate Dehydrogenase*
IPT	*Immunophenotyping*
ITD	*Internal tandem duplications*
JM	*Juxta-membrane*
LAIP	*Leukemia-associated immunophenotyped*
LDH	*Lactate dehydrogenase*
MDS	*Myelodysplastic syndrome*
MGB	*Minor groove binder*
mIDH	*Mutant IDH*
MPO	*Myeloperoxidase*
MRD	*Minimal Residual Disease*
Na	*Sodium*
NCCN	*National Comprehensive Cancer Network*
NFQ	*Nonfluorescent quencher dye*
NGS	*Next-generation sequencing*
NK	*Normal karyotype*
NOS AML	*Not otherwise specified AML*
NPM	*Nucleophosmin*
NSE	*Non- specific esterase*
OR	*Odds ratio*
OS	*Overall survival*
PAS	*Periodic acid- Schiff*
PE	*Phycoerythrin*
PR	*Partial remission*
R132	*Residue at codon 132*

List of Abbreviations cont...

Abb.	Full term
R132C	*Cysteine*
R132H	*Histidine*
RFS	*relapse-free survival*
ROC	*Receiver-operating characteristic*
RR	*Relative risk*
RT-PCR	*Reverse transcription-polymerase chain reaction*
RT-qPCR	*Real time quantitative PCR*
RUNX1	*Runt-Related Transcription Factor*
SBB	*Sudan black B*
SD	*Standard deviation*
SNP	*Single nucleotide polymorphism*
SRSF2	*Serine/arginine-rich- splicing-factor-2*
TdT	*Terminal deoxynucleotidyl transferase*
TET2	*Ten–Eleven Translocation 2*
TKD	*Tyrosine kinase domain*
TLC	*Total leucocytic count.*
Tm	*Meltingtemperature*
TN	*True negative*
TP	*True positive*
TP53	*Tumor Protein p53*
TRM	*Treatment-related mortality*
TSS	*Transcriptional start sites*
WHO	*World Health Organization*
WT	*Wilms-tumor*
α-KG	*α-ketoglutarate*

INTRODUCTION

Acute myeloid leukemia (AML) is a clonal malignant disease of hematopoietic tissue caused by somatic mutations in genes that control normal cell proliferation and differentiation ***(Paschka et al., 2010).***

The molecular genetic alterations are one of the most important prognostic factors that have been identified in AML and the role of these genetic alterations has been emphasized by the 2008 revised World Health Organization classification of AML like nucleophosmin (NPM) 1, and CCAAT enhancer-binding protein α (CEBPA), Wilms-tumor (WT1), Fms-like tyrosine kinase3 (FLT3) ***(Sjöblom et al., 2006).***

Identification of new gene mutations provides useful markers for diagnosis, prognosis assessment and making therapeutic decision with monitoring therapy ***(Scholl et al., 2009).*** Among these are epigenetic mutations that include isocitrate dehydrogenase mutations; IDH1 and IDH2 ***(Clark et al., 2016).***

IDH proteins are homodimeric enzymes involved in diverse cellular processes, including adaptation to hypoxia, histone demethylation and DNA modification ***(Clark et al., 2016).***

The IDH2 protein is localized in the mitochondria and is a critical component of the tricarboxylic acid (also called the 'citric acid' or Krebs) cycle. Both IDH2 and IDH1 (localized in

the cytoplasm) proteins catalyze the oxidative decarboxylation of isocitrate to α-ketoglutarate (α-KG) ***(Molenaar et al., 2014).***

The mutations confer neomorphic enzyme activity through the NADPH-dependent reduction of the normal end-product α-ketoglutarate to the putative oncometabolite 2-hydroxyglutarate. The accumulation of high levels of 2-hydroxyglutarate in the *IDH1/2*-mutant tumor provides an important mechanism of cellular transformation through the targeting of epigenetic regulators ***(Platt et al., 2015).***

The IDH1 and IDH2 mutations have been identified in glioma, cartilaginous tumors, thyroid carcinomas, cholangiocarcinoma, prostate cancers, paragangliomas, melanoma, chronic-, fibrotic-, or blast-phases of essential thrombocythemia, polycythemia vera or myelofibrosis, and AML. In AML, the IDH1 and IDH2 mutations are frequently associated with blastic transformation or aggressive forms ***(Nomdedéu et al., 2012).***

Testing for IDH is straightforward, given that nearly all IDH mutations are located on exon 4, and affect IDH1 at a single residue, Arg132, or IDH2 at two residues, Arg140 and Arg172 ***(Yang et al., 2012).***

Several methods, including PCR and sequencing, are commonly used for IDH detection ***(Mahdieh and Rabbani, 2013).*** Because IDH mutations occur in approximately one in

five patients with AML, mutational testing should be part of routine molecular assessment at diagnosis to identify patients who may in time benefit from targeted treatments currently under clinical study ***(Aref et al., 2015).*** Identification of these mutations at diagnosis may also be pivotal for better risk stratification of AML patients ***(Jin et al., 2014).***

AIM OF THE WORK

To evaluate the prognostic impact of both IDH1 and IDH2 mutations in newly diagnosed AML patients and their correlation with different clinical and laboratory parameters.