Effect of Zinc and Vitamin D on T Helper 17 Cells in Chronic HCV Patients

Thesis

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List of Abbreviations

Abb.	Full term
AHR	*Aryl Hydrocarbon Receptor*
AIH	*Autoimmune Hepatitis*
ALD	*Autoimmune Liver Disease*
ALT	*Alanine Transaminase*
ASC	*Asymptomatic Carrier*
BMI	*Body Mass Index*
cAMP	*Cyclic Adenosine Monophosphate*
CLD	*Chronic Liver Disease*
CMV	*Cytomegalovirus*
CO2	*Carbon Dioxide*
Cu	*Copper*
CXCL	*Chemokine*
EBV	*Epstein–Barr Virus*
EIA	*Enzyme Immunoassay*
ELISA	*Enzyme-Linked Immunosorbent Assay*
FICZ	*Formylindolo[3, 2-b]Carbazole*
G-CSF	*Granulocytes Colony-Stimulating Factor*
GM-CSF	*Granulocytes Macrophage Colony-Stimulating Factor*
HBV	*Hepatitis B Virus*
HCC	*Hepatocellular Carcinoma*
HCV	*Hepatitis C Virus*
HIV	*Human Immunodeficiency Virus*
HIV/AIDS	*Human Immunodeficiency Virus/Acquired Immune Deficiency Syndrome*
HSCs	*Hepatic Stellate Cells*
Ig	*Immunoglobulin*
IL	*Interleukin*
IO	*Ionomycin*

List of Abbreviations (Cont...)

Abb.	Full term
IR	*Immunoresponse*
IRF4	*Interferon Regulatory Factor 4*
KC	*Kupffer Cells*
KIR	*Killer Cell Immunoglobulin-Like Receptors*
MCP-1	*Monocyte Chemoattractant Protein-1*
MTF	*Metal-Regulatory Transcription Factor*
NADPH	*Nicotinamide Adenine Dinucleotide Phosphate*
NK	*Natural Killer*
PBMCs	*Peripheral Blood Mononuclear Cells*
PBS	*Phosphate Buffered Saline*
Peg-IFN	*Pegylated-Interferon*
Peg-RBV	*Pegylated- Ribavirin*
PMA	*Phorbol 12-Myristate 13-Acetate*
PGE-2	*Prostaglandin E2*
PSC	*Primary Sclerosing Cholangitis*
RDA	*Recommended Dietary Allowance*
RdRp	*RNA-Dependent RNA Polymerase*
RNA	*Ribonucleic Acid*
RNS	*Reactive Nitrogen*
ROS	*Reactive Oxygen Species*
RPMI	*Park Memorial Institute*
RUNX1	*Runt-Related Transcriptional Factor 1*
RXR	*Partner, The Retinoid X Receptor*
Se	*Selenium*
STAT3	*Signal Transducer and Activator of Transcription 3*
SVR	*Sustained Virologic Response*
TCA	*Trichloroacetic Acid*
TF	*Transcription Factor*

List of Abbreviations (Cont...)

Abb.	Full term
TGFβ	*transforming Growth Factor β*
Th	*T Helper*
TNF	*Tumor Necrosis Factor*
VDR	*Vitamin D Receptor*
VDREs	*Vitamin D Response Elements*
Zn	*Deficiency*
Zn	*Zinc*

ABSTRACT

Serum levels of Zn, Vit. D, and IL-17 were assessed in the patients group and subgroups. Patients lymphocytes were activated in vitro in the presence or absence of Zn or Vit.D3 in 2 different concentrations (low and high), and then intracellular IL-17 production was assessed using flow cytometry.

Our results showed that Zn and Vit. D were significantly decreased in HCV patients. Increasing disease severity leads to more reduction in Zn level opposed by increasing IL-17 level.

Zn potently reduced IL-17 production in a dose-related fashion. Vit. D apparently increases IL17 expression, it is unclear whether it is due to its toxic effect on cell count or lack of definite association between Vit. D and both IL-17 and disease severity.

Keywords: Partner, The Retinoid X Receptor - Signal Transducer and Activator of Transcription 3 - Sustained Virologic Response

INTRODUCTION

Hepatitis C is the disease that has affected around 200 million people globally. HCV is a life threatening human pathogen, not only because of its high prevalence but also because of the potentially serious complications of its persistence ***(Bostan and Mahmood, 2010).***

The outcome of HCV infection is determined within six months of exposure to the virus. Control of acute primary viral replication is associated with expansion of antiviral $CD4^+$ (helper) and $CD8^+$ (cytotoxic) T cells ***(David and Christopher, 2005).***

Th17 cells, named for their ability to produce interleukin-17A (IL-17A; also known as IL-17), are a novel subset of $CD4^+$ effector T helper (Th) cells. Th17 cells also produce IL-17F, as well as other cytokines and chemokines, including IL-21, IL-22, IL-26, and CCL20 ***(Pelletier et al., 2010).***

In human liver disease, there is strong evidence for the involvement of Th17 cells in a variety of inflammatory processes in the liver, including all major disease entities such as viral hepatitis. In HCV infections, reports indicate a close correlation between virus-induced liver inflammation, infiltration and activation of Th17 cells and the amount of liver damage caused by the antiviral immune response ***(Hammerich et al., 2010).***

Essential micronutrients are involved in many metabolic pathways in the liver; such as enzymatic functions, protein synthesis, oxidative reactions and anti-oxidant defense ***(Ko et al., 2005).***

Since the metabolism of micronutrients takes place in the liver, their concentrations may be varied with different types of liver disease. The levels of some essential micronutrients such as Zn, Cu, Fe and Se might exacerbate liver disease in case of deficiency, imbalance, or toxicity. Clinical studies reported that hepatitis C virus related chronic liver disease patients at different stages of liver damage have impaired metabolism of micronutrients ***(Mohammed et al., 2012).***

Zinc affects multiple aspects of the immune system. It is essential for normal development and function of cell-mediated innate immunity. Neutrophils, natural killer cells and macrophages are also affected by zinc deficiency. The growth and function of T and B cells are also affected adversely due to zinc deficiency ***(Prasad, 2009)***.

It is also well known that zinc is involved in cytoprotection of hepatocytes against oxidative stress. Serum zinc levels become reduced in association with the progression of chronic liver disease ***(Himoto et al., 2007)***.

Beside its effect on calcium and bone homeostasis, vitamin D is emerging as a critical factor involved in the regulation of the immune system, inflammatory response, and fibrogenesis. Recent findings in genotype 1 HCV patients have shown correlations between low serum levels of 25-OH vitamin D3 [25(OH)D] and severe lesions of liver fibrosis, and low sustained virologic response (SVR) to pegylated-interferon (Peg-IFN)-based therapy ***(Petta et al., 2010).***

Increased circulating Th17, intrahepatic IL-17 positive cells, as well as HCV-specific Th17 cells were positively correlated with severity of liver inflammation in chronic HCV patients ***(Bălănescu et al., 2012).***

Serum zinc levels become reduced in association with the progression of chronic liver disease ***(Himoto et al., 2007),*** also the prevalence of severe vitamin D deficiency increases with increasing severity of liver dysfunction ***(Kitson and Roberts, 2012).*** Researchers have claimed that zinc ***(Kitabayashi et al., 2010)*** and vitamin D ***(Chang et al., 2010)*** affect T helper 17 cells negatively. Yet, no enough research has been done on the effect of zinc and vitamin D on T helper17 cells in chronic HCV patients.

Aim of the Work

The aim of the work is to detect the in vitro effect of zinc and vitamin D on stimulated T helper 17 cells obtained from chronic HCV patients.