Study of HCV status in Egyptian cases of B non-Hodgkin's lymphoma with assessment of patients' immunological state

Thesis

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List of abbreviations

- ACD: Anemia of chronic disease
- ADCC: Antibody dependent cell-mediated cytotoxicty
- AHA: Autoimmune hemolytic anemia
- AID: Activation-induced cytidine deaminase
- ALCL: Anaplastic large cell lymphoma
- ALL: Acute lymphoblastic leukemia [
- alloSCT: Allogeneic stem cell transplantation
- ALP: Alkaline phosphatase
- ALT: Alanine transaminase
- ANA: Anti nuclear antibody
- ASCT: Autologus stem sell transplantation
- AST: Aspartate transaminase
- AT: Antiviral therapy
- BAFF: B-cell-activating factor
- BCA-1: B-cell-attracting chemokine-1
- BCR: B-cell receptor
- BL: Burkitt's lymphoma
- BLC: B-lymphocyte chemoattractant
- Bregs: Regulatory B cells
- BTK: Bruton's tyrosine kinase
- CALLA: Common acute lymphoblastic leukemia antigen
- CDC: Complement dependent cytotoxicity
- CDC: Complement dependent cytotoxicity
- CLDN1: Claudin-1
- CLL: Chronic lymphocytic leukemia.
- CMT: Combined modality therapy
- CODOX-M: Cyclophosphamide, vincristine, doxorubicin, and methotrexate
- COO: Cell-of-origin

- CR: Complete remission
- CT: Computed tomography
- DAAs: Direct-acting antivirals
- DA-EPOCH-R: Dose-adjusted etoposide, prednisone, vincristine, cyclophosphamide, doxorubicin, rituximab
- DHAP: Dexamethasone, AraC, cisplatin
- DLBCL: Diffuse large B-cell lymphoma
- DRC: R-dexamethasone-cyclophosphamide
- EBV: Epstein-Barr virus
- ELISA: Enzyme linked immunosorbantassay
- FL: Follicular-Lymphoma
- FLIPI: Follicular-Lymphoma International Prognostic Index
- GAG: Glycosaminoglycans
- GCB: Germinal center B cells
- G-CSF: Granulocyte colony-stimulating factor
- GDP: Gemcitabine, dexamethaxone, cisplatin
- GP: Glycoprotein
- HAART: Highly active retroviral therapy
- HCL: Hairy cell leukemia
- HCV: Hepatitis C virus
- HDC: High dose chemotherapy.
- HD-MTX: High-dose methotrexate
- HL: Hodgkin lymphoma
- HyperCVAD: Hyperfractionated cyclophosphamide, vincristine, doxorubicin, and dexamethasone
- IARC: International Agency for Research on Cancer
- ICE: Ifosfamide, carboplatin, etoposide
- IFRT: Involved-field radiotherapy
- IgA: Immunoglobulin A
- IgD: Immunoglobulin D
- IgE: Immunoglobulin E

- IgG: Immunoglobulin G
- IgM : Immunoglobulin M
- Igs; Immunoglobulins
- IHC: Immunohistochemical
- IPI : International Prognostic Index
- IRES: Internal ribosome entry site
- IT: Intrathecal
- ITP: Idiopathic thrombocytopenia purpura
- IVAC: Ifosfamide, etoposide, and cytarabine
- kDa: KiloDalton
- KSHV: Kaposi sarcoma–associated herpesvirus
- LDH: Lactate dehydrogenase.
- LDLR: low-density lipoprotein receptor
- LPDs: lymphoproliferative disorders
- LPL : Lymphoplasmacytic lymphoma
- MALT: Mucosa-associated lymphoid tissue ()
- MC: Mixed cryoglobulinemia
- MCL: Mantle Cell Lymphoma
- MG: Monoclonal gammopathies
- MGUS: Monoclonal gammopathy of undetermined significance
- MIPI: Mantle Cell Lymphoma International Prognostic Index
- MiRNA: MicroRNA
- MM: Multiple Myloma
- MRI: Magnetic resonance imaging
- MZL: Marginal zone B-cell lymphomas
- NCCN: The National Comprehensive Cancer Network
- NHL: B-cell non-Hodgkin's lymphoma
- NNPIs: NS5B non-nucleoside polymerase inhibitors
- NPIs: Nucleoside polymerase inhibitors
- NS: Nonstructural
- OPN: Osteopontin
- PBMCs: Peripheral blood mononuclear cells

- PCNSL: Primary CNS lymphoma
- PCR: Polymerase chain reaction
- PIs: Protease inhibitors
- PMBCL: Primary mediastinal (thymic) large B-cell lymphoma
- PS: Performance status
- R2CdA: R-2-chloro-2'-deoxyadenosine
-
- R-ACVBP: Adriamycin, cyclophosphamide, vindesine, bleomycin, and prednisone
- R-CHOP: Rituximab, cyclophosphamide, vincristine, prednisone
- R-CVP: Rituximab, cyclophosphamide,prednisone
- REAL: Revised European-American Lymphoma
- RF: Rheumatoid factor ()
- R-IPI: International Prognostic Index revised IPI
- rNTP: Ribonucleoside triphosphates
- RTX: Rituximab
- SLL: Small lymphocytic leukemia.
- SMZL: Splenic marginal zone lymphoma
- SR-BI: Scavenger receptor class B type I,
- SVR: Sustained virological response

- TdT: Terminal deoxynucleotidyl transferase
- TTP: Thrombotic Thrombocytopenic Purpura
- V/D/J segments: variable (V), diversity (D) and joining (J) segments
- WBC: White blood cell
- WHO: World Health Organization
- WM: Waldenström macroglobulinemia

Abstract

Hepatitis C virus (HCV) infection has been associated with the development of B-cell non-Hodgkin lymphoma (including diffuse large B-cell lymphoma, marginal zone lymphoma, lymphoplasmacytic lymphoma, and extranodal marginal zone B cell lymphoma of mucosa-associated lymphoid tissue) as well as primary hepatic lymphoma . In addition, HCV infection may increase the risk of hepatotoxicity related to treatment for lymphoma.

The risk of NHL was increased in patients with HCV compared with non-HCV infected individuals.

Lymphoma may develop due to the progression of cryoglobulinemia.
One hypothesis is that cryoglobulinemia arises from chronic stimulation of the immune system by HCV, predisposing to a lymphoproliferative disorder.

Some data suggest that successful treatment of HCV may reduce the risk of lymphoma in patients who achieve a sustained virologic response.

Key words:
Hepatitis C virus, lymphoproliferative disorder, B cell non-Hodgkin's lymphoma, Cryoglobulinemia.

Introduction and aim of the work

Hepatitis C virus (HCV), a hepatotropic and lymphotropic virus is frequently associated with benign and malignant lymphoproliferative disorders such as mixed cryoglobulinaemia and B-cell non-Hodgkin's lymphoma (NHL) including diffuse large B-cell lymphoma, marginal zone lymphoma, lymphoplasmacytic lymphoma, and extranodal marginal zone B cell lymphoma of mucosa-associated lymphoid tissue) as well as primary hepatic lymphoma **(Monti et al.,2005).**

Findings that support an association of HCV with lymphoma include:
The prevalence of HCV in patients with B-cell non-Hodgkin lymphoma (NHL) was much higher than the prevalence in the general population or in patients with other hematologic malignancies **(Ennishi et al., 2010).**

The risk of NHL was increased in patients with HCV compared with non-HCV infected individuals **(Monti et al., 2005).**

The development of unexplained anemia or lymphadenopathy in a patient with HCV and clinically active cryoglobulinemia should raise concern about an underlying lymphoproliferative disorder **(Ennishi et al., 2010).**

Some data suggest that successful treatment of HCV may reduce the risk of lymphoma in patients who achieve a sustained virologic response**(Cocoub et al., 2000).**

Whether treatment of the underlying HCV infection could be effective in patients who have developed lymphoma is uncertain. Regression of splenic lymphoma has been described in association with HCV treatment **(Quinn et al., 2001**).

Some data suggest that successful treatment of HCV may reduce the risk of lymphoma in patients who achieve a sustained virologic response **(Marignani et al., 2011).**

In this work we aimed to study HCV status in cases diagnosed as B non-Hodgkin's lymphoma and the characteristics of non-Hodgkin's lymphoma in HCV positive patients with assessment of their immunological state through serum immunoglobulins assay.

The hepatitis C virus

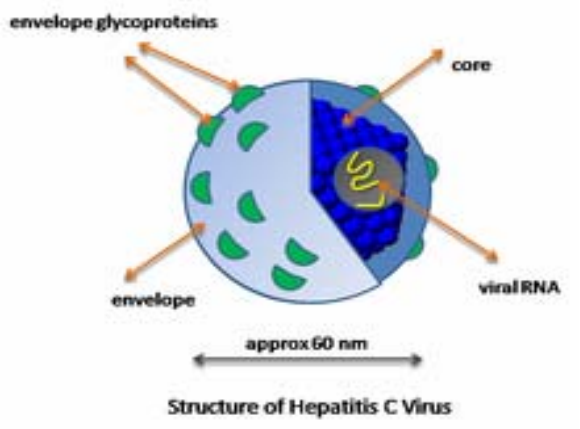
The hepatitis C virus is a single stranded, enveloped, positive sense RNA virus.

Taxonomy

The hepatitis C virus belongs to the genus Hepacivirus a member of the family Flaviviridae **(Op De Beeck et al.,2003).**

Structure

The hepatitis C virus particle consists of a core of genetic material (RNA), surrounded by an icosahedral protective shell of protein, and further encased in a lipid (fatty) envelope of cellular origin. Two viral envelope glycoproteins, E1 and E2, are embedded in the lipid envelope **(Jubin R 2001).**



FigureI: Simplified diagram of the structure of the Hepatitis C virus particle

Genome

Hepatitis C virus has a positive sense single-stranded RNA genome. The genome consists of a single open reading frame that is 9600 nucleotide bases long. This single open reading frame is translated to

produce a single protein product, which is then further processed to produce smaller active proteins **(Berry et al., 2011).**

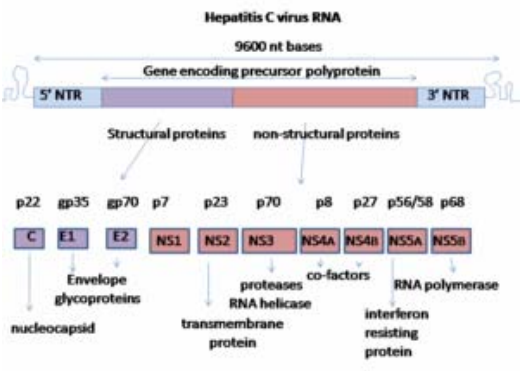


Figure II: Genome organization of Hepatitis C virus

At the 5' and 3' ends of the RNA are the UTR that are not translated into proteins but are important to translation and replication of the viral RNA. The 5' UTR has a ribosome binding site (IRES — Internal ribosome entry site) that starts the translation of a very long protein containing about 3,000 amino acids. The core domain of the hepatitis C virus (HCV) IRES contains a four-way helical junction that is integrated within a predicted pseudoknot **(Dubuisson J, 2007).**

The conformation of this core domain constrains the open reading frame's orientation for positioning on the 40S ribosomal subunit. The large pre-protein is later cut by cellular and viral proteases into the 10 smaller proteins that allow viral replication within the host cell, or assemble into the mature viral particles **(Gupta G et al., 2012).**

Structural proteins made by the hepatitis C virus include Core protein, E1 and E2; nonstructural proteins include NS2, NS3, NS4A, NS4B, NS5A, and NS5B.

Molecular biology

The proteins of this virus are arranged along the genome in the following order: N terminal-core-envelope (E1)–E2–p7-nonstructural

protein 2 (NS2)–NS3–NS4A–NS4B–NS5A–NS5B–C terminal. The mature nonstructural proteins (NS2 to NS5B) generation relies on the activity of viral proteinases **(Jin et al., 2012).**

The NS2/NS3 junction is cleaved by a metal dependent autocatalytic proteinase encoded within NS2 and the N-terminus of NS3. The remaining cleavages downstream from this site are catalysed by a serine proteinase also contained within the N-terminal region of NS **(Baghbani-arani et al., 2012).**

The core protein has 191 amino acids and can be divided into three domains on the basis of hydrophobicity: domain 1 (residues 1–117) contains mainly basic residues with two short hydrophobic regions; domain 2 (resides 118–174) is less basic and more hydrophobic and its C-terminus is at the end of p21; domain 3 (residues 175–191) is highly hydrophobic and acts as a signal sequence for E1 envelope protein. Both envelope proteins (E1 and E2) are highly glycosylated and important in cell entry. E1 serves as the fusogenic subunit and E2 acts as the receptor binding protein. E1 has 4–5 N-linked glycans and E2 has 11 N-glycosylation sites **(Kohaar et al., 2010).**

The p7 protein is dispensable for viral genome replication but plays a critical role in virus morphogenesis. This protein is a 63 amino acid membrane spanning protein which locates itself in the endoplasmic reticulum. Cleavage of p7 is mediated by the endoplasmic reticulum's signal peptidases. Two transmembrane domains of p7 are connected by a cytoplasmic loop and are oriented towards the endoplasmic reticulum's lumen **(Zeisel et al., 2009).**

NS2 protein is a 21–23 kDa transmembrane protein with protease activity.

NS3 is 67-kDa protein whose N-terminal has serine protease activity and whose C-terminal has NTPase/helicase activity. It is located within