Section A-Research paper

An Overview about SEN and Torque Teno Viruses Among Hemodialysis Patients

Haytham Kamal Ahmed Mahrous [1], Hisham M. Omar[1], Ezzat Mostafa Mohamed [2], Mohamed Essam Tawfik Atwa[1], Manal M. El Gerby[1]

1 Department of Clinical Pathology, Faculty of Medicine, Zagazig University
2 Department of Internal Medicine, Faculty of Medicine, Zagazig University
Email: Midoessam83@gmail.com

Article History: Received 10th June, Accepted 5th July, published online 10th July 2023

Abstract

Background: SEN virus is considered one of the blood-borne viruses. It was discovered by investigators at DiaSorin Biomolecular Research Institute, Saluggia, Italy in their work for searching about viral causes of post transfusion hepatitis which is not due to the hepatitis C virus or hepatitis B virus. These cases have been called "non-A-to-E hepatitis" because they are not due to any other of the well-known human hepatitis viruses. SEN-V has considered one of the most commonly viral infection found in hemodialysis patients (HD). HD patients are prone to acquire parentally transmitted viral infections such as Hepatitis B, Hepatitis C and HIV. They considered a high risk group for acquiring infection by blood borne viruses because the HD materials which have been used in various medical centers are not completely disposable in addition to the fact that the therapeutic interventions are mostly associated with bleeding and blood transfusion. TT virus has been discovered in 1997 in Japan from the bloodstream of patients with post-transfusion hepatitis, and its name referred to the initials of the first patient in whom virus was diagnosed. The name can also indicate a transfusion-transmitted virus; despite transfusions are not the only means of transmission. After that this virus was called Torque Teno, keeping the initials TTV. In a study carried out on dialysis patients in Italy, The prevalence of TTV DNA in dialysis patients was significantly greater (41.7%) than in healthy individuals (10.7%). Concurrent infection with HCV among dialysis patients positive for TTV has been reported. On the other hand, Low TTV frequency and no correlation between TTV and other blood-borne infections have been reported in study on HD patients in Iran, also TTV infection was not associated with high levels of liver enzymes.

Keywords: SEN Virus, Torque Teno Virus, Hemodialysis

DOI:- 10.53555/ecb/2023.12.Si12.254

Introduction

In medicine, hemodialysis is a method for removing waste products such as potassium and urea, as well as free water from the blood when the kidneys are in renal failure. Hemodialysis is one of three renal replacement therapies (the other two being renal transplant; peritoneal dialysis). Hemodialysis can be an outpatient or inpatient therapy. Less frequently hemodialysis is done at home. Dialysis treatments in a clinic are initiated and managed by specialized staff made up of nurses and technicians; dialysis treatments at home can be self initiated and managed or done jointly with the assistance of a trained helper who is usually a family member **(1).**

The principle of hemodialysis is the same as other methods of dialysis; it involves diffusion of solutes across a semipermeable membrane. Hemodialysis utilizes counter current flow, where the dialysate is flowing in the opposite direction to blood flow in the extracorporeal circuit. Countercurrent flow maintains the concentration gradient across the membrane at a maximum and increases the efficiency of the dialysis. Fluid removal

(ultrafiltration) is achieved by altering the hydrostatic pressure of the dialysate compartment, causing free water and some dissolved solutes to move across the membrane along a created pressure gradient **(2).**

The dialysis solution that is used is a sterilized solution of mineral ions. Urea and other waste products, and also, potassium and phosphate, diffuse into the dialysis solution. However, concentrations of sodium and chloride are similar to those of normal plasma to prevent loss. Bicarbonate is added in a higher concentration than plasma to correct blood acidity. A small amount of glucose is also commonly used **(2).**

Side-effects and complications

There are symptoms resulting from fluid removal, such as low blood pressure, fatigue, chest pains, leg cramps, nausea, and headaches, collectively known as the "dialysis hangover." The severity of these side effects varies and can be managed by adjusting fluid intake and treatment frequency **(3).**

Hemodialysis may expose their circulatory system to microbes, which can lead to sepsis, an infection affecting the heart valves (endocarditis) or an infection affecting the bones (osteomyelitis). The risk of infection varies depending on the type of access used. Bleeding may also occur. Infections can be minimized by strictly adhering to infection control best practices **(3).**

First Use Syndrome is a rare but severe anaphylactic reaction to the artificial kidney. Its symptoms include sneezing, wheezing, shortness of breath, back pain, chest pain, or sudden death. It can be caused by residual sterilant in the artificial kidney or the material of the membrane itself. Long-term complications of hemodialysis include amyloidosis, neuropathy and various forms of heart disease. Increasing the frequency and length of treatments have been shown to improve fluid overload and enlargement of the heart that is commonly seen in such patients **(4).**

SEN virus is considered one of the blood-borne viruses. It was discovered by investigators at DiaSorin Biomolecular Research Institute, Saluggia, Italy in their work for searching about viral causes of post transfusion hepatitis which is not due to the hepatitis C virus or hepatitis B virus. These cases have been called “non-A-to-E hepatitis” because they are not due to any other of the well-known human hepatitis viruses **(5).**

Nomenclature

The name of SEN-V refers to the initials of the infected patient from whom the SEN virus was first isolated. He was injection drug user infected with a human immunodeficiency virus. The first publication about the nucleic acid sequence of SEN virus was done on 18 May, 2000. After that reports of other studies by other researchers have been done in 2001 **(6)**.

Structure

Recently, a new virus, designated SEN virus (SENV), was identified as a putative non-A to E hepatitis virus. This virus contains a single-stranded circular DNA of 3900 nucleotides in length and is distantly related to TTV **(**7**).**

The genome was formed from 3,900 nucleotides and at least three open reading frames (ORF). The ORF 1 with Arg/Lys-rich domains is the largest one with hydrophilic characteristic. The function of ORF2 is unknown. The ORF3 translation results in forming a protein with similarity to a DNA topoisomerase I so, ORF3 seems to have an important role in the replication of the virus **(8)**.

SEN-V earlier belonged to the family circoviradae, genus Anellovirus after that changes in nomenclature have classified anelloviruses capable of causing human infection. SEN virus has a high mutation rate, so it is more similar to RNA viruses rather than DNA viruses and this characteristic feature gave the virus the ability of persistence **(9)**.

Genetic Variation

Nine genotypes (A-I) have been detected for SEN-V; among which genotypes D and H are more frequent in individuals infected with hepatitis of unknown origin compared to other genotypes **(10).**

The majority of research papers have reported on D and H genotypes as clinically significant strains. They are the only factors detected in patients with unknown hepatitis (non-A to E hepatitis) and healthy blood donors but less commonly **(10).**

Epidemiology

Today, SENV has a global incidence with variable occurance geographically. It is the latest viral agent that has been proposed as a cause of non-A-G hepatitis **(11).**

The prevalence of SEN-virus has been increased in association with HIV-1 and hepatitis C and B viruses, also injection drug users have shown high level of infection with this virus. This association suggests that the parenteral route is considered the most common route of transmission **(12).**

Many studies were done to define the frequency of SENV-D and SENV-H in transfused people. Thalassemic patients and healthy blood donors in Iran, that high prevalence of SEN-V infection among thalassemic patients indicates blood transfusion as the main route of transmission, also high percentage of SEN-V infection among healthy individuals suggests that there are other methods which are important in transmission of SEN-V **(13).**

Iatrogenic means in hospital setting represent another method for transmission of SEN-V. Also, it can be transmitted from mother to fetus (vertical transmission). It was found that the distribution of SEN-V in hepatitis A patients is higher than in healthy adults. This suggests fecal-oral transmission as possible route of SEN-V transmission. Also, TTV has been detected in stool from some cases. The close relationship between SEN-V and TTV supports the idea of SEN-V fecal-oral transmission **(14)**.

SEN-V and Liver Diseases

Different studies have discussed the importance, pathogenesis and clinical presentation of SEN-V infection and the association between SEN-V infection and development of liver diseases. Levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), total bilirubin; alkaline phosphatase (ALP), gamma-glutamyl transpeptidase and also the pathological picture of patients who were positive for SEN-V in comparison to negative patients were reported. But, these studies showed that there was no evidence suggests that SEN-V causes hepatitis when it is the sole agent detected or even worsen the severity or progression of coexistent chronic HCV **(15).**

Persistent infection by SEN-V may extend over one year and were reported in some cases as long as 12 years **(9)**.

The role of SENV infection and the clinical significance have been studied in patients with non A-E hepatitis or other viral hepatitis but the results are not very obvious and are inconsistent to some extent and even show contradictory results. That is to say, co infection of SEN-V with hepatitis B virus, or hepatitis C virus were found to be associated with severe and fulminant liver diseases. On the other side, other studies have reported that SEN-V has no well-established pathogenicity and the role of this virus in development of liver diseases as acute and chronic hepatitis, cirrhosis and the development of hepatocellular carcinoma still needs to be confirmed **(16)**.

Moreover, others have suggested that SEN-V has a protective role against HCV **(9)**.

Hosseini and Bouzari, 2016 reported that SEN-V was found in 90% of the healthy individuals, 66% of HBV positive individuals, and 46% of HCV-positive individuals. The prevalence of SEN-V especially its two genotypes (D&H) were significantly lower in HBV and HCV patients ($P<0.01$) than in healthy individuals. Also, the frequency of SENV-H was higher than SENV-D in all studied groups. The level of liver enzymes (ALT and AST) in HBV patients who were SEN-V positive were significantly less than negative cases ($P<0.05$). The same finding was observed for SENV-H-negative and positive patients, this suggests a positive impact of the virus in hepatic pathogenicity by lowering hepatic damage and thus decreasing the liver enzymes**.** Although SEN-V has been found in high frequency in the serum of patients with hepatocellular carcinoma however, there is no well-established evidence that incriminates SEN-V in the development of HCC **(5).**

SEN-V among patients on maintenance hemodialysis

SEN-V has considered one of the most commonly viral infection found in hemodialysis patients (HD). HD patients are prone to acquire parentally transmitted viral infections such as Hepatitis B, Hepatitis C and HIV. They considered a high risk group for acquiring infection by blood borne viruses because the HD materials which have been used in various medical centers are not completely disposable in addition to the fact that the therapeutic interventions are mostly associated with bleeding and blood transfusion **(17)**.

SEN-V co-infection

Hepatitis C virus and hepatitis B virus have been incriminated in development of liver pathology. SEN-V infection is more frequent in patients with chronic HBV or HCV. Co-infection with SEN-V was frequently reported in 22-67% of chronic HBV and 20-76% of HCV patients **(12).**

Co-infections with SEN-V and HIV-1 were found in Italy, the United States, and Germany. Individuals who acquired HIV via use of intravenous drugs showed higher Frequency of SEN-V (71%) than those who acquired HIV by sexual transmission (26%). These results may suggest the blood-borne transmission of SEN-V. HIV-1 infection also may probably facilitate maternal-fetal transmission of SEN-V **(5).**

Diagnosis of SEN-V infection

SEN-V has been detected by using

The DNA enzyme immunoassay (EIA) method. In this method, viral DNA is amplified by polymerase chain reaction and detected using biotinylated strain-specific probes in an EIA.

Also, SEN-V is detected using either PCR-Southern blot or ethidium bromide gel electrophoresis after single-step PCR **(18).**

Seminested PCR or nested PCR. The assays show high specificity. Some or all of the amplified SEN-V amplicons have been sequenced in some of the studies **(5).**

Torque Teno virus (TTV)

TT virus has been discovered in 1997 in Japan from the bloodstream of patients with post-transfusion hepatitis, and its name referred to the initials of the first patient in whom virus was diagnosed. The name can also indicate a transfusion-transmitted virus; despite transfusions are not the only means of transmission. After that this virus was called Torque Teno, keeping the initials TTV **(19).**

Structure

At first, sequence of 500 nucleotides (nt) has been documented and then this sequence extended to up to 3700 nt. Sequence analysis of viral genome at that time lead to suggestion that TTV has been related to the Parvoviridae family. Two other different studies reported the presence of an additional GC-rich region of about 120 nucleotide (nt) at the end of 1998. After that the circular nature of the TTV genome was discovered. This finding has confirmed the relationship of TTV with the Circoviridae family **(20).**

TTV genome is a circular single-stranded DNA molecule of negative polarity. It contains a GC-rich region formed of 117 nucleotides. The TTV genome has been divided into an untranslated region (UTR) of 1.2 kb and another coding region of 2.6 kb. The UTR region is considered to some extent conserved region, leading to suggestion that it has an essential regulatory role in replication of virus. On the other side the coding region of anelloviruses has two large open reading frames: ORF1 and ORF2.After that many other open reading frames have been documented, and the proteins that they encode vary in length for different isolates **(19)**.

Genetic Variation of TTV

While TTV bears some similarity to members of the group Circoviridae, it lacks sequence homology with any known viruses, and it is believed to be the first known member of a new family of viruses. It is classified under the Anelloviridae family, Alpha torque teno virus genus. Many other closely related TTV sequences were discovered after discovery of TTV. They are genetically grouped in about twenty nine different species. This reflects a very high degree of genetic heterogeneity, to a degree close to that present in RNA viruses **(20)**.

TTV genome is very diverse in nature. Until now there are well defined sixty one different isolates worldwide. The reasons and mechanisms for such great genomic diversity are not clear up till now.Although, it has been reported that parvoviruses with single stranded DNA genomes mutate at high rates, and it was suggested that this high rate of mutation may be illustrated by the nature of the single stranded genome and its ability to encode peptides involved in replication process. This explains why TTV mutates at a high rate **(21)**.

Also there are many other possible causes that explain higher than expected level of mutation that occur in TTV. It may be due to Intra genomic rearrangement, presence of hyper variable regions in open reading frame, or due to Recombination **(21).**

More than thirty genotypes of TTV were classified into five groups. The genotypes 1, 2, 3 and 4 seem to be frequently distributed all over the world, but the distribution of genotype 5 is not completely documented .

Another Few subtypes (1a, 1b, 2a, 2b) have also been defined. The genogroups are uniquely present in different area all over the world with G1 and G2 TTV more frequent in the USA and in Italy in patients with liver disorders and in blood donors. On the other side, G4 is also identified but is less frequent. Many of these genotypes are reported to be incriminated in human diseases **(22)**.

TTV-Like Viruses and Relatives of TTV

After TTV has been discovered, other three types of TTV-like viruses were also found. They are called: Torque Teno Mini virus (TTMV), Small Anellovirus (SAV) and Torque Teno Midi virus (TTMDV) **(23).**

1. **Torque teno mini virus (TTMV)**

   TTMV is characterized by having smaller genome size and length than TTV. It accidently has been found in samples from human plasma during PCR reaction using specific primers for TTV that primarily base paired with the sequences and produced a shorter segment than TTV **(24)**.

2. **Small anellovirus (SAV)**

   Small anellovirus also has some homology with TTV. It has a GC-rich segment, a coding region and a Chicken Anemia Virus (CAV) like motif .SAV isolates form a large phylogenetic tree and is frequently distributed over the world among healthy individuals **(25)**.

3. **Torque teno midi virus(TTMDV)**

   Since short time during the amplification of SAV genome from human sera the amplicons obtained have been longer than expected and their length was 3242-3253 nt and they show all the characters of TTV like viruses. These newly identified amplicons were known as TTMDV. After study the various TTMDV genome sequences it has been documented that they contain a large group of isolates that have various lengths and sequence. As regard to nucleotide sequence it shows 31% variability and at amino acid level it shows 61% variability **(26)**.

4. **Resemblance of TTV with Chicken Anemia Virus (CAV)**

   CAV belongs to *Circoviridae* family. This family characterized by single stranded DNA genome and the ability to infect vertebrates. Gyrovirus and Circovirus are considered two genus of *Circoviridae*. CAV belongs to genus circovirus which is a pathogen of chicken; it shows homology with TTV regarding organization of genome and regulatory sites for transcription. Both TTV and CAV have negative sense circular ss DNA genome **(23).**

   In addition, TTV shows similarity with CAV in having arginine rich N-terminus within ORF1 which has essential role in binding and assembly of viral DNA within the capsid. Another homology between both TTV and CAV is that TTV derived apoptosis inducing protein is similar to the apoptin protein of CAV that causes apoptosis in cancer cells **(23).**

5. **Resemblance of TTV with Hepatitis C Virus**

   TTV genotype 1a has ORF3 which codes for a protein. This protein shows homology with non-structural protein 5A (NS5A) of HCV and has essential role in inhibiting the Interferon-induced anti-viral response (**27)**.

   TTV concurrent infection with HCV were studied all over world, However it is still needed to establish whether TTV is considered an "accidental visitor", "helper" or "responsible cause" for hepatic pathology caused by HCV **(28)**.

6. **Animal Torque Teno Virus**

TTV has not been found in human only but it was also detected in the sera of non-human primates and domesticated farm animals. TTV is frequently detected among the animals worldwide with different genome length and sequence. The TTV of animals often has smaller genome than human TTV **(29).**

Epidemiology

TTV is distributed worldwide affecting different age groups **(30)**.

The highest TTV distribution was detected in thalassemic patients receiving multiple transfusions, hemodialysed patients for long time, hemophiliacs receiving clotting factor, and intravenous drug users. The high TTV load may refer to the immunological status of the individual as immunosuppressed individuals show great viral titers **(19)**.

TTV DNA has also been found in 14.9 percent of hospital surfaces and 16 % of air samples which were contaminated via using toilet. The researchers reported that TTV can be used as an indicator of general viral contamination of the surroundings **(19)**.

Route of transmission

TTV has been found in plasma, peripheral blood mononuclear cells, various body fluids and secretions like saliva, tears, semen, vaginal fluid, breast milk and stools. TTV also was detected in other body organs such as kidneys, prostate, brain, mammary glands, and bone marrow cells (BMCs). TTV has parenteral route for transmission via transfusion of blood and blood products, and it is known to be shed with the bile into the feces of infected patients. This suggests that it may be transmitted via fecal-oral route **(30)**.

The relatively high distribution of TTV among blood donors in addition to the majority of TTV infected patients with no history of transfusion of blood and blood products point to other routes of transmission of TTV infection. Some studies documented placental route for transmission of TTV, however others have not found TTV in cord blood and amniotic fluid. Transmission via sexual route is of low frequency **(20).**

Comparison of TTV sequence from both mothers and their offspring showed both homology and difference. So that, TTV has been acquired by children via more than one route either from the mother or from the surroundings **(19)**.

Pathogenesis

It was documented that TTV is found in both the nucleus and cytoplasm of some of the peripheral blood mononuclear cells (PBMCs). TTV infection of cells of immune system may play a role in evasion of the virus from the immune system**.** TTV in PBMCs may act as a reservoir of TTV for chronicity of the infection, and also transmission in some clinical cases and epidemiological distribution. In vivo, TTV shows a high ability to replicate. More than 1010 TTV virions have been produced daily; this has been concluded from the kinetics of viral load and clearance during interferon therapy **(31)**.

The definitive way by which TTV replicates is not well understood. Due to similarities of TTV to other single stranded DNA-genome viruses, it is suggested that TTV replicates by using the rolling-circle replication mechanism. ORF1 was known to encode peptide that includes replication motives **(31)**.

There was a hypothesis suggested that TTV may replicate by using an RNA intermediate, such as HBV, However it seems different as the TTV genome does not have the ability to encode a reverse transcriptase. Due to diversity of genome of TTV, this also led to suggest another hypothesis that viral DNA is replicated by mechanism with poor or absent proofreading activity **(19)**.

As regard to the tissue tropism and cell-specificity of TTV, several studies have reported that TTV does not have a special tissue tropism, and virus can replicate in many tissues/organs such as (liver, spleen, bone marrow, lymph nodes, muscles, pancreas, lungs, thyroid and kidneys). However, viral titers have been up to 10–300 times higher in specific tissues as (bone marrow, spleen, lung, and, liver) than in the serum of the same individual **(19)**.

TTV among patients on maintenance hemodialysis

In a study carried out on dialysis patients in Italy, The prevalence of TTV DNA in dialysis patients was significantly greater (41.7%) than in healthy individuals (10.7%). Concurrent infection with HCV among dialysis patients positive for TTV has been reported **(32)**.

On the other hand, Low TTV frequency and no correlation between TTV and other blood-borne infections have been reported in study on HD patients in Iran, also TTV infection was not associated with high levels of liver enzymes **(32)**.

TTV as a Marker of Immune Status

Replication of TTV virus is controlled by the immunological status of individuals who harbor it (higher viral loads have been found in individuals with immunodeficiencies, like AIDS patient and other with intercurrent diseases) **(33)**.

Large distribution of TTV reflects a strong interaction between it and the host. Researchers cannot know how effectively immune system withstands TTV infection and controls super infections by other different species of the virus. TTV produces a microRNA in vivo that interferes with action of interferons and thus microRNA

plays an essential role in evasion of virus from immune attack by interference with the host antiviral response and directly participates in the high distribution of these viruses **(33)**.

In patients with lymphoma or myeloma and receiving high-dose chemotherapy with autologous hematopoietic stem cell transplantation, the level of TTV viraemia was inversely associated with the percentage of CD8+ 57+ T lymphocytes which are indicators of immune competent state. In addition, the slope of return from peak to baseline TTV values has been found to be indicative of the time required for a patient to recover immune competence before having the next dose of chemotherapy **(34)**.

Patients who undergo solid organ transplantation show marked changes in TTV level, and these changes in TTV kinetics are the same regardless of the type of transplanted organ. In children and adult undergo liver transplantation and receive maintenance immunosuppression therapy, it was found that TTV load correlates with the intensity of immunosuppression **(34)**.

Diagnosis of TTV

Nucleic acid-based detection:

The vast genetic diversity, which presumably leads to high antigenic diversity, and the difficulty in obtaining representative control antigens and antibodies introduce significant variability in the detection of TTVs, which is primarily achieved by nucleic acid detection methods. Assays targeting conserved UTR sequences enable the detection of a majority of TTVs, while those targeting more variable areas of the genome help to distinguish between genera.

Several conventional or quantitative PCR (qPCR) based assays, rolling circle amplification and in situ hybridization have been reliably tested. As PCR based methods are limited by the range of the primers used, they may not be versatile enough to capture the complete diversity within a sample. To improve the accuracy of detection and accommodate the genetic diversity of the family, it is recommended that a panel of primer/ probe combinations be used for PCR-based assays, especially in the absence of whole genome sequencing .While nucleic acid-based detection is versatile and critical for observational science, using a combination of nucleic acid and antigen-based assays such as immunohistochemistry can improve clarity in assigning biological significance to diagnostic findings **(35).**

Serological detection:

Enzyme linked immunosorbent assays (ELISAs) based on the ORF1 are available for TTV1a, 1b. An ORF2 based ELISA is available for TTV1a. Generally, the presence of antibodies is well correlated with qPCR-based detection, with an increase in antibody titers reducing viral loads **(34)**.

Difficulties in collecting much information about TTV were due to a shortage of essential diagnostic tools, such as efficient in vitro culture systems and sensitive serologic methods to identify and investigate viral antigens and antibodies. In addition, TTV was first detected at a time when HIV and hepatitis viruses attracted the attention and resources which were available to researchers. Thus, investigation of TTV remained for long time limited to certain laboratories over the world. However, information to date collected by many investigators has been enough to create scientific interest about the virus and the interaction with its host **(34)**.

PCR techniques have undergone substantial evolution. Detection of TTV can be done using universal primer, which amplifies most, if not all, the human TTVs, or using specific primer, which allows sorting of the virus in one of the 29 species into which TTVs were subdivided. Nowadays, real time PCR technique with SYBR Green based quantification assay for identifying human TTV titers from plasma were reported **(36)**.

References

1. Murdeshwar HN, Anjum F. Hemodialysis. [Updated 2023 Apr 27]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024 Jan-.
2. Oshvandi K, Kavyannejad R, Borzuo SR, Gholyaf M. High-flux and low-flux membranes: efficacy in hemodialysis. *Nurs Midwifery Stud*. 2014;3(3):e21764.
3. Languluri R, Shaik Ayub B, Kanala S K. Dialysis Treatment: A Comprehensive Description, ijpras,2014,3(1)1-13.
4. Daugirdas JT, Ing TS. First-use reactions during hemodialysis: a definition of subtypes. *Kidney Int Suppl*.

1988;24:S37-S43.

5. Akiba J, Umemura T, Alter HJ, Kojiro M, and Tabor E (2005): SEN virus: epidemiology and characteristics of a transfusion-transmitted virus. TRANSFUSION; 45:1084-1088.
6. Mohamed IS, Thabit AG, Abd-El Rahman SA, Mohammed EA and Seif Eldin SS (2011): Prevalence of SEN Virus Infection in Multitransfused Patients in Assiut University Hospitals, Egypt. Journal of American Science; 7(1):687-698.
7. Joshua S. Davis, Ginger Chu & Prabuddhua Pathinayake, et al.,.,2020): Seroprevalence of Torque Teno Virus in hemodialysis and renal transplant patients in Australia, Wiley Periodicals LLC, 2020;22:e13400.
8. Tanaka Y, Hayashi J, Ariyama I, Furusyo N, Etoh Y, Kashiwagi S (2000). Seroepidemiology of TT virus infection and relationship between genotype and liver damage. Digestive diseases and sciences 45:2214-2220.
9. Umemura T, Alter HJ, Tanaka E, Orii K, Yeo AE, Shih JW, Matsumoto A, Yoshizawa K and Kiyosawa Km (2002): SEN virus: response to interferon alfa and influence on the severity and treatment response of coexistent hepatitis C. Hepatology; 35:953–959.
10. Abbasi S., Makvandi M., Karimi G., Neisi N. The prevalence of SEN virus and occult hepatitis B (OBI) virus infection among blood donors in Ahvaz City. Jundishapur J. Microbiol. 2016;9:3–8.
11. Shymaa A. Elaskary, Doaa S. Elgendy & Fathia Elbassal et al.,.,2023): Prevalence of SEN Virus and Torque Teno Virus in Hemodialysis Patients and Healthy Blood Donors in Menoufia University Hospitals, Egyptian Journal of Medical Microbiology, January 2023;32(1):141-150.
12. Mohammad SM, Hamideh SM and Moayed SA (2009): Is SEN Virus a Major Concern in Hemodialysis and Liver Transplantation? Hepatitis Monthly; (9):50-59.
13. Karimi-Rastehkenari A and Bouzari M (2010): High frequency of SEN virus infection in thalassemic patients and healthy blood donors in Iran.Virology Journal;7:1.
14. Serin MS, Koksal F , Oksuz M , Abayli B, Aslan G, Tezcan S, Yildiz C, Kayar B and Emekdas G (2005): SEN virus prevalence among non-B and non-C hepatitis patients with high liver function tests in the south of Turkey. J pn J Infect Dis.; 58(6):349-352.
15. Omar M, Saad El-Din S, Fam N, Diab M, Shemis M, Raafat M, Seyam M , Hassan M, Badawy F, Akl M and Saber M (2008): SEN Virus Infection in Egyptian Patients With Chronic Hepatitis C and Patients Undergoing Hemodialysis. Medscape J Med.; 10(12): 290.
16. Elsherbiny NM, Hassan EA, Ahmed AO, Abd El-Rehim AS, Abd El Rehim N and Hassany SM (2015): Does SEN virus (SENV) infection affect the progression of chronic hepatitis C or B among Egyptian patients? African journal of microbiology research; 9(23):1504-1512.
17. Nazish M, Rizvi M, Azam M, Haque SF and Malik A (2015): A Study of Prevalence and Associated Risk Factors of SEN Virus in Haemodialysis Patients.Int.J.Curr.Microbiol.App.Sci.;1: 134-142.
18. Kao JH, Chen W, Chen PJ, Lai MY, and Chen DS (2003): SEN virus infection in patients with chronic hepatitis C: preferential coinfection with hepatitis C genotype 2a and no effect on response to therapy with interferon plus ribavirin. J Infect Dis; 187:307-10.
19. Yukari AT, Okubo P, Keiko PS, Haruki RY, Angélica M E, Spandole S, Cimponeriu D, Mariana LB, and Mihaescu G (2015): Human anelloviruses: an update of molecular, epidemiological and clinical aspects Arch Virol .;160:893–908.
20. Irshad M, Joshi YK, Sharma Y, and Dhar I (2006): Transfusion transmitted virus: A review on its molecular characteristics and role in medicine. World J Gastroenterol. ; 12(32): 5122–5134.
21. Shackelton LA, Parrish CR, and Holmes EC (2006): Evolutionary basis of codon usage and nucleotide composition bias in vertebrate DNA viruses. J Mol Evol.; 62:551–563.
22. Muratori P, Czaja AJ, Muratori L, et al. Genetic distinctions between autoimmune hepatitis in Italy and North America. World J Gastroenterol. 2005;11(12):1862-1866.
23. Bostan N, Amen NE and Bokhari H (2013): Current and Future Prospects of Torque Teno Virus, J Vaccines

Vaccin; 4:1.

24. Hsiao KL, Wang LY, Cheng JC, Cheng YJ, Lin CL, Liu HF. Detection and genetic characterization of the novel torque teno virus group 6 in Taiwanese general population. *R Soc Open Sci*. 2021;8(11):210938.
25. Biagini P, de Micco P, de Lamballerie X. Identification of a third member of the Anellovirus genus ("small anellovirus") in French blood donors. Arch Virol. 2006;151(2):405-408.
26. Ninomiya M, Takahashi M, Shimosegawa T and Okamoto H (2007): Analysis of the entire genomes of fifteen torque teno midi virus variants classifiable into a third group of genus Anellovirus. Arch Virol.; 152: 1961-1975.
27. Cordek DG, Bechtel JT, Maynard AT, Kazmierski WM, Cameron CE. TARGETING THE NS5A PROTEIN OF HCV: AN EMERGING OPTION. *Drugs Future*. 2011;36(9):691-711.
28. Marrie TJ, Tyrrell GJ, Majumdar SR, Eurich DT. Concurrent Infection with Hepatitis C Virus and Streptococcus pneumoniae. Emerg Infect Dis. 2017;23(7):1118-1123.
29. Okamoto H, Takahashi M, Nishizawa T, Tawara A , Fukai K Muramatsu U, Naito Y and Yoshikawa A (2002): Genomic characterization of TT viruses (TTVs) in pigs, cats and dogs and their relatedness with species-specific TTVs in primates and tupaias. J Gen Virol.; 83: 1291-1297.
30. Ghasemi PD, Doosti A and Hajimirzaei MR (2011): The Prevalence of Transfusion-Transmitted Virus (TTV) Infection in Iranian Patients with Chronic Hepatitis B. International Journal of Medical, Health, Biomedical, Bioengineering and Pharmaceutical Engineering; 5(2):65-68.
31. Maggi F, Pifferi M, Fornai C, Andreoli E, Tempestini E, Vatteroni M, Presciuttini S, Marchi S, Pietrobelli A, Boner A (2001). TT virus in the nasal secretions of children with acute respiratory diseases: relations to viremia and disease severity. Journal of Virology 77:2418-2425.
32. Rivanera D, Lozzi MA, Idili C, Lilli D. Prevalence of TT virus infection in Italian dialysis patients. Pathol Biol (Paris). 2009;57(1):97-100.
33. Zhong S, Yeo W, Tang MW, et al. Gross elevation of TT virus genome load in the peripheral blood mononuclear cells of cancer patients. Ann N Y Acad Sci. 2001;945:84-92.
34. Focosi D, Macera L, Boggi U, Nelli LC, Maggi F. Short-term kinetics of torque teno virus viraemia after induction immunosuppression confirm T lymphocytes as the main replication-competent cells. J Gen Virol. 2015;96(Pt 1):115-117.
35. Ssemadaali, M. A., K. Effertz, P. Singh, O. Kolyvushko and S. Ramamoorthy (2016). "Identification of heterologous Torque Teno Viruses in humans and swine." Sci Rep 6: 26655.
36. Garibyan L, Avashia N. Polymerase chain reaction. *J Invest Dermatol*. 2013;133(3):1-4.