Jpn. J. Infect. Dis., 54, 55-63, 2001

**Review**

# GB Virus-C/Hepatitis G Virus

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(Received January 25, 2001. Accepted April 23, 2001)

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**SUMMARY**: GB virus-C (GBV-C)/hepatitis G virus (HGV) is a positive, single-strand RNA virus that has been classified in the family *Flaviviridae*. Interestingly, GBV-C/HGV appears to have a truncated or absent core protein at the amino terminus of the polyprotein. GBV-C/HGV is transmitted parenterally and probably sexually. Most GBV-C/HGV infections appear to be asymptomatic, persistent, and no correlation between virus infection and liver dysfunction although the disease-inducing activity of GBV-C/HGV remains to be investigated. Furthermore, there was no evidence of pathogenesis in the liver by experiment with chimpanzees. From these results, GBV-C/HGV might be considered as a kind of "orphan" virus in search of a disease. Epidemiological investigation demonstrated that GBV-C/HGV infection is present in about 1-1.4% of the healthy population in developed countries and in 8-14.6% in developing countries. The genome of GBV-C/HGV exhibits a sequence variation among different isolates. On the basis of this variation, it has been proposed that GBV-C/HGV can be classified into at least four major genotypes, consisting of type 1 (West Africa), type 2 (US/Europe), type 3 (Asia), and type 4 (Southeast Asia).

## Introduction

After discovery of the hepatitis A and B viruses (HAV, HBV) in the 1960s and 1970s, it became apparent that an unidentified agent was responsible for hepatitis in a number of patients suspected of having acute or chronic viral hepatitis. The unknown pathogen was referred to as non-A, non-B hepatitis until the hepatitis C virus (HCV) was identified in 1989 (1) and was found to account for hepatitis in the majority of these patients. However, in approximately 10% of patients with liver diseases, no cause can be identified. It has long been suspected that additional hepatotropic viruses will be discovered to account for some of these cases, especially since approximately half of these patients have a history of blood transfusion. The cloning of the HCV established that it is among the most important human pathogens. In 1996, using similar methods, two new RNA viruses were independently discovered in human serum and were named GB virus-C (GBV-C) and hepatitis G virus (HGV), respectively (2,3). Database screening indicated that these viruses had high sequence similarity, hence they are now regarded as independent isolates of the same virus. This article will provide a review of the available information concerning GBV-C/HGV.

## History

GBV-C is the acronym for a virus originally derived from a surgeon (whose initials were G.B.) in whom hepatitis developed in the 1960s (4). His plasma caused hepatitis in tamarins, and the 11th passage in tamarin serum was the cloning source of the virus. Three different GBV agents have been cloned thus far; GBV-A and GBV-B appear to be tamarin viruses, and GBV-C is a human virus (2,5). GBV-C was discovered by gene amplification with primers derived from shared sequences in GBV-A, GBV-B, and HCV (2). On the other hand, the initial source from which HGV was cloned was a patient with community-acquired chronic hepatitis (3). The sequences of GBV-C and HGV are more than 95% homologous, and the two are considered to be closely related isolates of the same virus.

## Viral genome

The genomic organization of GBV-C/HGV is similar to that of HCV (Fig. 1). The GBV-C/HGV genome is a positive-

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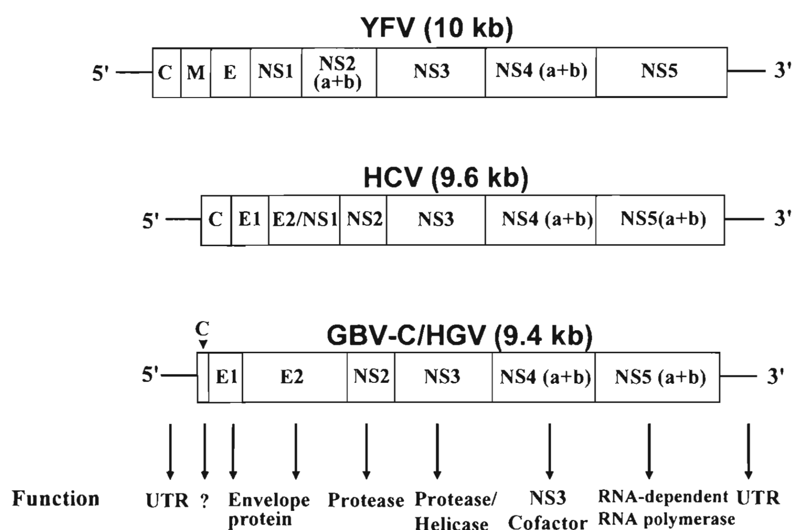


Fig. 1. Comparison of genome structure among yellow fever virus (YFV), HCV, and GBV-C/HGV. UTR, untranslated region; C, core gene; E1 and E2, envelope genes 1 and 2; NS2 through NS5b, non-structural genes 2 through 5b.

strand RNA of about 9400 nucleotides (nt) and it contains a large open reading frame that encodes a polyprotein precursor of about 2900 amino acid (aa) residues. Like HCV, GBV-C/HGV has characteristics of a flavivirus-like genome. The two viruses thus represent a new genus in the family *Flaviviridae* including flaviviruses and pestiviruses, since the genetic distance between GBV-C/HGV and HCV is too great to consider GBV-C/HGV as a different genotype of HCV (2). Like HCV, the genome of GBV-C/HGV is preceded by a 5′-untranslated region (UTR), which is followed by a long open reading frame. The genome terminates with a 3′ UTR, but it has no poly(A)-tail. In contrast to HCV, the sequence variability of envelope 2 (E2) is very low among different isolates collected worldwide and no hypervariable region exists in the envelope region. The presumed polyprotein contains structural envelope 1 (E1) and E2 glycoproteins at the amino-terminal end, followed by nonstructural (NS) proteins (NS2, NS3, NS4, NS5) at the carboxy-terminal end. The E2 glycoprotein has three potential glycosylation sites and 18 cysteine residues that might be involved in disulfide bonds. Interestingly, the GBV-C/HGV genome lacks a clearly identifiable core gene which encodes a nucleocapsid protein. In our study, from the size of the putative core region at the aa level in 54 GBV-C/HGV isolates, they can be divided into four different groups (unpublished data). In other words, 2 isolates with 107 aa (group 1) were found, 4 isolates with 84 aa (group 2), 9 isolates with 47 aa (group 3), and 39 isolates with 16 aa (group 4). In addition, one isolate lacked the core region completely. In contrast, the core region of HCV consists of 191 aa. The lack of a core-like protein at the N-terminus of the viral polyprotein distinguishes GBV-V/HGV from all other members of the *Flaviviridae*. Thus, GBV-C/HGV appears to be distinct from enveloped viruses in general because they tend to encode a basic protein that mediates the packaging of the viral nucleic acid into the virion envelope. However, Schmolke et al. (6) reported that GBV-C/HGV builds classic viral particles displaying E2 envelope protein on their outer surfaces by using a panel of eight monoclonal antibodies against the putative E2 protein following DNA immunization. In addition, Xiang et al. (7) reported that GBV-C/HGV particles included an extremely-low density virion particle (1.07 to 1.09 g/ml) and a nucleocapsid of ~1.18 g/ml, as shown on a sucrose gradient. Their data also suggested that plasma samples from individuals chronically infected with GBV-C/HGV contained antibody to the GBV-C/HGV core protein peptide. These data indicate that GBV-C/HGV has a nucleocapsid and that at least part of the putative core region of the virus is expressed in vivo. In the literature, infectious particles lacking such a core have been generated artificially with the vesicular stomatitis virus glycoprotein (8). The 5′UTR contains an internal ribosome entry site that is capable of directing CAP-independent translation of the polyprotein (9,10). Multiple sequence alignments demonstrated that the 5′ UTR and 3′ UTR contains blocks of highly conserved (>95%) sequences. This conservation has enabled the optimization of RT-PCR assays using primers from these conserved regions.

### Mutation rate of the viral genome

Nakao et al. (11) reported that GBV-C/HGV was estimated to have a mutation rate of $3.9 \times 10^{-4}$ base substitutions per site per year. We also determined the mutation rate of GBV-C/HGV. For this purpose, we isolated and compared two GBV-C/HGV genomes (HGV-IM68 isolate and HGV-IM71 isolate) from the same Japanese patient. The IM68 isolate had been obtained from the patient 12 years before the study took place. The IM68 sequence differed from IM71 sequence at 59 (1%) of the 5867 nt. The differences between those isolates were exclusively nucleotide point mutations. Based on these results, the mutation rate of HGV-IM71 was estimated to be $0.8 \times 10^{-3}$ base substitutions per site per year during a 12-year period. This result suggests that the mutation rate of the GBV-C/HGV genome is almost the same as that of HCV (12-14). However, unlike HCV, GBV-C/HGV does not have a hypervariable region in the entire gene. The highest rate of mis-sense mutation was found in the E2 region, which was followed by the rate of mutations in E1 and NS4 regions. No mis-sense mutation was seen in the core region, nor in the NS5b region. Moreover, no nucleotide mutation was observed in the 5′-UTR. Potential Asn-linked glycosylation (N-X-T/S) motifs located within the E1 and E2 regions were found to be conserved entirely between the HGV-IM71 and -IM68 isolates.

## Diagnostic assay

RT-PCR is the only diagnostic tool to detect GBV-C/HGV RNA. The nucleotide sequences in the 5'UTR and the 3'-UTR were highly conserved among different isolates of GBV-C/HGV isolates (Fig. 2). Thus, these regions are the most suitable for RT-PCR use. An enzyme immunoassay for the detection of antibodies to the E2 protein of GBV-C/HGV has been reported by several groups (15-17). The presence of GBV-C/HGV RNA in serum reflects active viral infection, but the presence of anti-E2 antibody has been reported to correlate with recovery from viral infection, because GBV-C/HGV E2 antibody was usually detected in individuals without viral RNA and E2 antibody appeared in association with the cessation of viremia. These results suggest that the presence of anti-E2 of GBV-C/HGV indicates past infection.

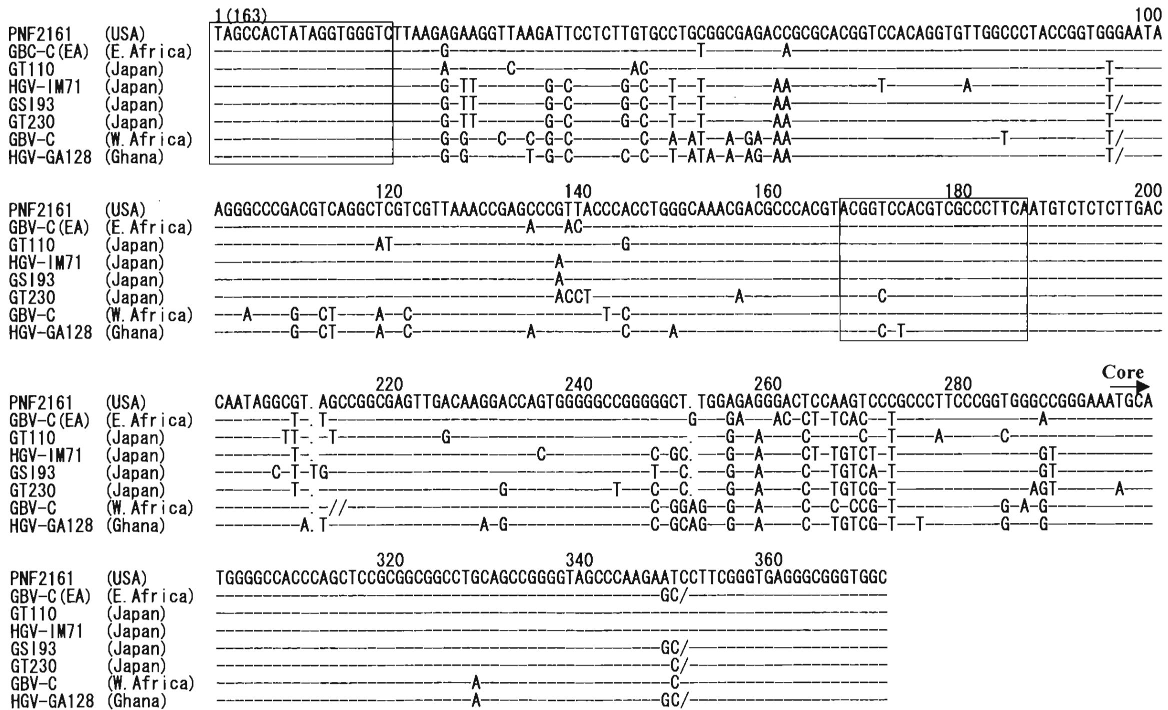

Fig. 2. Alignment of nucleotide sequences in the GBV-C/HGV 5'UTR (a) and the 3'UTR (b). Dashes indicate nucleotides identical to the top sequence. Sequences used for PCR primers are boxed.

## Epidemiology and clinical significance

Retrospective studies in our laboratory have revealed that GBV-C/HGV RNA has been found in the serum or plasma of volunteer blood donors at a frequency of 1-1.4% in developed countries and 8-14.6% in developing countries (with the exception of 1% in Nepal) (Table 1). GBV-C/HGV has a high prevalence among patients receiving hemodialysis treatment (6.2-26%), in patients who have received blood products, and in intravenous drug users (IVDU) (33-75%). Thus it seems likely that GBV-C/HGV is transmitted parenterally. In addition, the virus is common in patients who are also infected with other hepatitis viruses such as HBV (10%) and HCV (19%), perhaps because of similar modes of transmission. Although GBV-C/HGV RNA has been detected in 1-1.4% of healthy individuals and in 5-52% of liver disease patients (3,18-20), the role of GBV-C/HGV infection in liver diseases is not well understood. It appears that persistent infection with this virus is common, but the data do not support an association with significant liver injuries (21-23). GBV-C/HGV has also been seen in 7-50% of patients with non-A-E acute or fulminant hepatitis (18,22,24,25), but its causal role has not been established. In the Japanese population, GBV-C/HGV RNA was detected in 45 of 663 (6.8%) serum samples from patients with liver disease (Table 2), and it was found in 2 of 145 (1.4%) healthy individuals (18). Among the liver disease patients, GBV-C/HGV RNA was present in 4 of 58 (6.9%) with acute hepatitis, 16 of 229 (7%) with chronic hepatitis, 5 of 58 (8.6%) with liver cirrhosis, and in 17 of 139 (12.2%) with hepatocellular carcinoma. On the other hand, GBV-C/HGV RNA was also detected in 3 of 175 (1.7%) patients with non-viral liver diseases, including alcoholic liver disease, fatty liver, autoimmune hepatitis, primary biliary cirrhosis and drug-induced hepatitis. We were unable to detect the GBV-C/HGV genome within the livers of 54 patients with cryptogenic liver diseases (26). Among 45 HGV-positive patients, GBV-C/HGV coinfection with HBV and/or HCV were observed in 4 (8.9%) and 30 (66.7%), respectively, and in one (2.2%) patient with a triple infection involving these viruses (Table 2). Ten (22.2%) of 45 HGV-positive patients were positive for GBV-C/HGV alone. However, there was no significant difference discovered among alanine aminotransferase levels (ALT) in chronic hepatitis patients who were positive for GBV-C/HGV+HCV and HCV alone. Masuko et al. (21) reported that among GBV-C/HGV-positive blood donors and hemodialysis patients, there was no correlation between GBV-C/HGV RNA levels and elevation in serum ALT. Furthermore, no evidence has demonstrated that GBV-C/HGV acquired during liver transplantation contributes to post-operative liver function test abnormalities or has any impact on clinical progress. Neither age nor sex of patients was correlated to the prevalence of GBV-C/HGV infection in our study. Only 14 of 45 (31%) patients infected with GBV-C/HGV had a history of blood transfusions (18). The remaining 31 of 45 (69%) GBV-C/HGV-positive patients had no history of blood transfusions nor intravenous drug abuse. Mother-to-infant transmission of GBV-C/HGV does occur and the rate of transmission is much higher than that of HCV (27-29). GBV-C/HGV infection in newborns induce persistent viremia, but the infection is not accompanied by any symptoms of liver disease.

Table 1. Prevalence of GBV-C/HGV among healthy populations in various countries

| Country | $n$ | Positive for GBV-C/HGV RNA (%) |
|---|---|---|
| Japan | 145 | 2 (1.4) |
| Korea | 120 | 2 (1.2) |
| U.S. | 102 | 1 (1) |
| Spain | 104 | 1 (1) |
| Nepal | 397 | 4 (1) |
| Myanmar | 213 | 17 (8) |
| Vietnam | 242 | 28 (12) |
| Bolivia | 574 | 84 (14.6) |
| Egypt | 82 | 9 (11) |
| Ghana | 30 | 3 (10) |

## Hepatotropism and extrahepatic replication of the virus

In order to understand the pathogenesis in the liver, it is important to clarify whether or not GBV-C/HGV is a hepatotropic virus. GBV-C/HGV genome organization was found to be similar to that of HCV. Considering this genomic resemblance, it can be assumed that GBV-C/HGV replicates similarly to HCV, i.e., through negative-strand RNA, the presence of which could be regarded as direct evidence of viral replication. Based on this theory, Laskus et al. (30) have shown that all livers obtained from 10 patients who were seropositive for GBV-C/HGV were negative for the presence

Table 2. Prevalence of HGV infection in patients with various liver diseases in Japan (1985-1996)

| Disease | No. of subjects | HGV total (%) | HGV alone (%) | Co-infected with (%) | | |
|---|---|---|---|---|---|---|
| | | | | HBV | HCV | HBV+HCV |
| Acute hepatitis | 58 | 4 (6.9) | 4/4 (100) | 0 | 0 | 0 |
| Fulminant hepatitis | 4 | 0 | 0 | 0 | 0 | 0 |
| Chronic hepatitis | 229 | 16 (7.0) | 0 | 3/16 (18.8) | 13/16 (81.2) | 0 |
| Liver cirrhosis | 58 | 5 (8.6) | 1/5 (20) | 1/5 (20) | 2/5 (40) | 1/5 (20) |
| Hepatocellular carcinoma | 139 | 17 (12.2) | 2/17 (11.8) | 0 | 15/17 (88.2) | 0 |
| Alcoholic liver disease | 50 | 2 (4) | 2/2 (100) | 0 | 0 | 0 |
| Autoimmune hepatitis | 21 | 0 | 0 | 0 | 0 | 0 |
| Primary biliary cirrhosis | 27 | 0 | 0 | 0 | 0 | 0 |
| Fatty liver | 67 | 1 (1.5) | 1/1 (100) | 0 | 0 | 0 |
| Drug-induced | 10 | 0 | 0 | 0 | 0 | 0 |
| Total | 663 | 45 (6.8) | 10/45 (22.2) | 4/45 (8.9) | 30/45 (66.7) | 1/45 (2.2) |

(ref. 18)

of the GBV-C/HGV-RNA negative strand and only six were positive for the presence of the positive strand. In striking contrast, negative and positive strands of HCV RNA were detected in the liver specimens from HCV-seropositive patients. We also obtained the same results by PCR using liver tissue (data not shown). These findings imply that the liver is not the primary replication site for GBV-C/HGV. Absence of replication in liver tissue may explain the lack of influence of GBV-C/HGV on the progression toward liver disease. In contrast, Madejon et al. (31) and Saito et al. (32) have shown indirectly that GBV-C/HGV can replicate in the liver. The genomic GBV-C/HGV strand has been detected in peripheral blood mononuclear cells (PBMC). In addition, Fogeda et al. (33) have shown the in vitro infection of PBMC by GBV-C/HGV. Recently, they reported the heterogeneity and quasispecies composition of GBV-C/HGV isolated from in vitro-infected PBMC and from sera, livers, and PBMC from chronically infected patients. Their report showed the existence of different GBV-C/HGV variants with various levels of tissue tropism (34). As regards diseases associated with viral infection, Ellenrieder et al. (35) have demonstrated an increased prevalence of GBV-C/HGV (16.3%) in patients with low-grade non-Hodgkin's lymphoma. Further studies to evaluate the clinical and pathological significance of GBV-C/HGV infection in PBMC will be needed in order to clinically useful conclusions.

### Relation to hepatocellular carcinoma occurrence

Hepatocarcinogenicity is another important key question with regard to this virus. To elucidate the prevalence and association of GBV-C/HGV infection among hepatocellular carcinoma patients, we carried out a retrospective study by in situ detection of HBV, HCV, and GBV-C/HGV genome in 265 hepatocellular carcinoma tissues from various countries (36). HBV DNA and HCV RNA were detected frequently in carcinoma tissues. However, GBV-C/HGV RNA was undetected in all tested specimens, despite the inclusion of liver specimens from 5 GBV-C/HGV-seropositive patients. This suggests that GBV-C/HGV does not play an important role in hepatocarcinogenesis.

### Experimental infection in chimpanzees

It is known that chimpanzees are susceptible to GBV-C/HGV infection. Bukh et al. (37) reported that the infection of GBV-C/HGV in experimentally infected chimpanzees was characterized by the late appearance (weeks 10 and 11 after inoculation) of viremia that persisted throughout a 120-week follow-up. However, despite the relatively high titer of viremia, none of the chimpanzees developed hepatitis. This result suggests that persistence of GBV-C/HGV and HCV might occur by different mechanisms. HCV has a hypervariable region in the E2 protein that may circumvent the immune system and permit viral persistence. On the other hand, a hypervariable region in GBV-C/HGV was not identified.

### Co-infection in human immunodeficiency virus (HIV)-infected patients

High frequency of exposure to HBV and HCV is seen in HIV-infected subjects. To clarify the infection rate of GBV-C/HGV in HIV-infected patients, we investigated the prevalence of GBV-C/HGV, HBV, and HCV infections among 258 patients who were seropositive for anti-HIV antibody. The serum samples were collected from patients in Southeast Asia and in the United States (U.S.). The results revealed that GBV-C/HGV RNA was detected in 74 (29%) of 258 patients (Table 3). On the other hand, 66 (26%) and 29 (11%) patients were positive for HCV RNA and HBsAg, respectively. Among these patients, sexual contact was a more significant predictor of risk of GBV-C/HGV than HCV infection (Table 4). Lefrere et al. (38) and Yeo et al. (39) reported that patients with GBV-C/HGV infection have higher CD4 T cell counts and carriage of this virus is associated with a slower progression of HIV disease in coinfected patients. Further careful studies are required to confirm these results.

### Existence of viral genotypes and its geographic distribution

Based on variation in nucleotide sequence of 5′-UTR, GBV-C/HGV can be classified into three major genotypes.

Table 3. Prevalence of HGV, HCV, and HBV in HIV-infected patients

| Country | Tested number | HGV RNA | HCV RNA | HBsAg | G+C | G+B | B+C | B+C+G |
|---|---|---|---|---|---|---|---|---|
| Vietnam | 62 | 19 (31) | 17 (27) | 6 (10) | 5 (8) | 2 (3) | 0 | 0 |
| Myanmar | 89 | 25 (28) | 29 (33) | 15 (17) | 10 (11) | 3 (3) | 2 (2) | 0 |
| Cambodia | 9 | 4 (44) | 0 | 1 (11) | 0 | 1 (11) | 0 | 0 |
| Thailand | 26 | 3 (12) | 0 | 3 (12) | 0 | 0 | 0 | 0 |
| U.S. | 72 | 23 (32) | 20 (28) | 4 (6) | 4 (6) | 1 (1) | 0 | 1 (1) |
| Total | 258 | 74 (29) | 66 (26) | 29 (11) | 19 (7) | 7 (3) | 2 (0.7) | 1 (0.4) |

Number in parenthesis indicate percentages.

Table 4. Risk category with HCV, HGV, and HBV infections among HIV-infected patients in Southeast Asia

| Risk category | HCV ($n$=46) | HGV ($n$=51) | HBV ($n$=25) |
|---|---|---|---|
| Heterosexual transmission | 4 (9%) | 13 (25%) | 6 (24%) |
| Commercial sex workers | 4 (9%) | 18 (35%) | 5 (20%) |
| Intravenous drug users | 37 (80%) | 16 (31%) | 10 (40%) |
| Unknown | 1 (2%) | 4 (10%) | 4 (16%) |

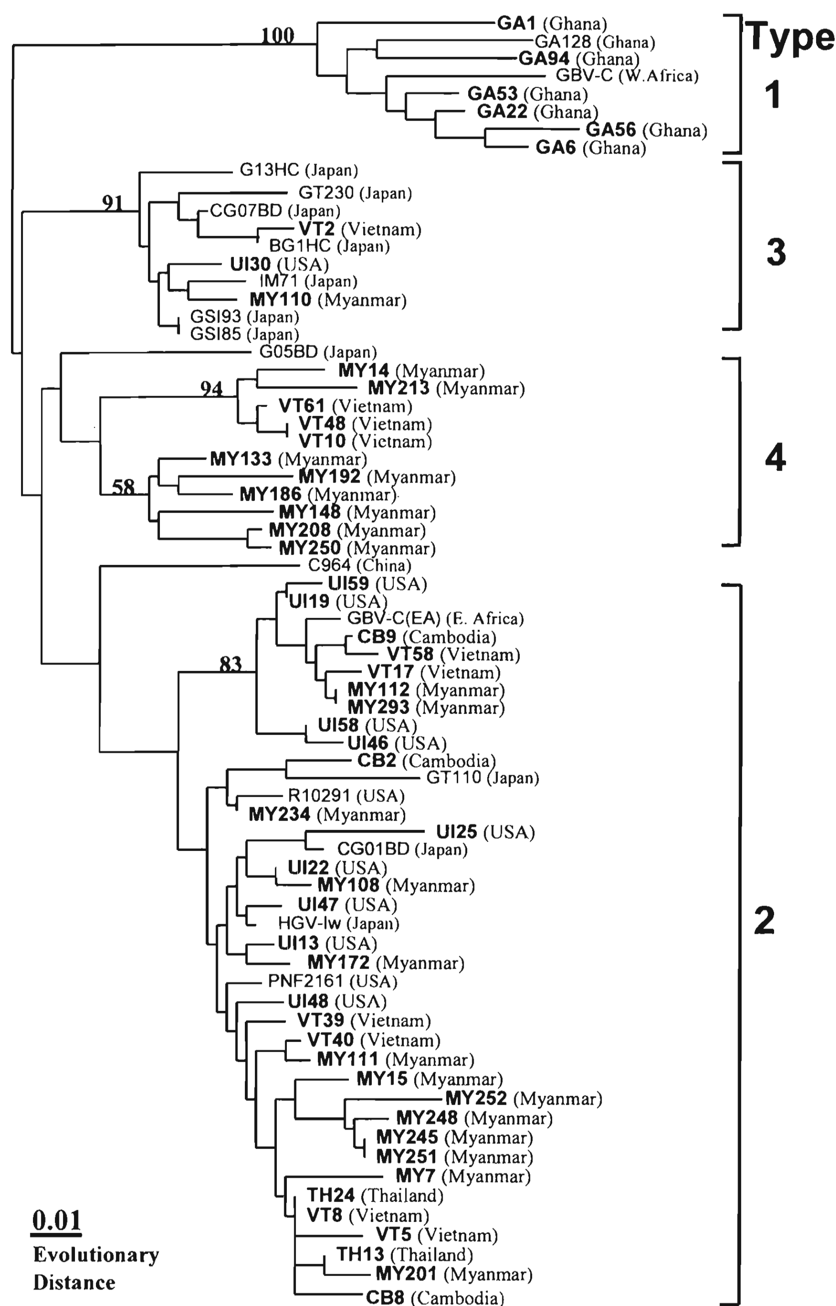


Fig. 3. Phylogram generated by neighbor-joining analysis of genetic distances in the 5′UTR of HGV/GBV-C isolates. Database-derived GBV-C/HGV isolates are presented in thin lettering. The percentage of bootstrap replicates supporting these branches is shown by the number given.

In addition to this classification, we recently identified a novel genotype of GBV-C/HGV in Southeast Asia and designated it as type 4 (Fig. 3) (40,41). In order to allow for the determination of genotypes 1, 2, 3, and 4 of GBV-C/HGV, we developed a simple and precise genotyping system based on PCR using type-specific primers (42). The type-specific primers were designed on the basis of the conserved nature of those sequences within a genotype, and of their poor homology with the sequences derived from other GBV-C/HGV genotypes. The alignment of the sequences of PCR primers used in our PCR genotyping system is shown in Fig. 4. By this new method, the frequency of infection with the four classified major genotypes of GBV-C/HGV was investigated in 251 infected individuals and/or patients from the following 12 different countries: Ghana, U.S., Spain, Egypt, Nepal, Thailand, Myanmar, Vietnam, Cambodia, Japan, the Philippines, and Bolivia. The results revealed that type 1 was detected in 28 of 251 (11%) serum samples, mainly these from Ghana (Table 5). Type 2 was distributed in 10 out of 12 countries investigated, but mainly in the U.S., Spain, Egypt, Nepal, and Myanmar. Type 3 was also distributed in various countries, but it was mainly in Japan and Bolivia. Type 4 was prevalent mainly in Vietnam, Myanmar, and Cambodia. Four of the 251 samples (1.6%) did not yield any specific products by PCR genotyping, hence the samples were considered unclassified. In Bolivia, our epidemiological study revealed that GBV-C/HGV RNA was present in 79 out of 574 (13.8%) blood donors, whereas HBsAg was detected in only 2 (0.3%), and no individuals

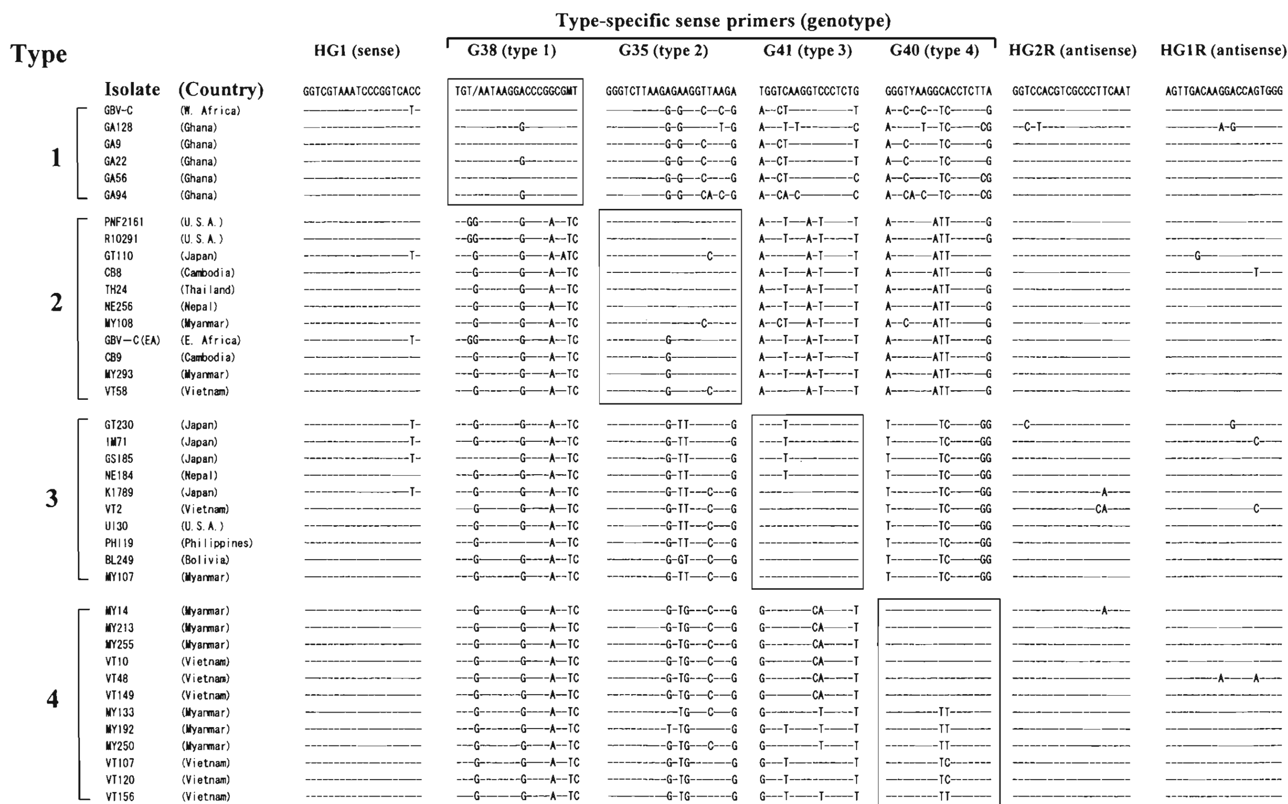

Fig. 4. Alignment of nucleotide sequences in the 5′UTR of GBV-C/HGV isolates and the positions of universal and type-specific primers. A total of 39 isolates from databases were compared to design the sequence of the PCR primers. Conserved sequences within each genotype are boxed. Dashes indicate nucleotides identical to the top sequence used for the PCR primers. Conserved sequences within each genotype are shown in boxes. (ref. 42)

positive for HCV RNA were found (43). Interestingly, we found that the major genotype of GBV-C/HGV in infected Bolivians is type 3, i.e., the Asian type, followed by type 2. This result may be correlated to the translocation of Mongoloid migrants who migrated from Asia to South America, including Bolivia, a long time ago. In fact, people of Mongoloid descent account for more than half of the general population in Bolivia. Similar genotypic distributions are seen in the molecular epidemiology of human T-cell leukemia virus (44).

## Conclusion

An understanding of the biology and clinical significance of GBV-C/HGV is evolving. Increasing evidence suggests that GBV-C/HGV is not a significant cause of liver disease, although it may be capable of inducing hepatitis in a subset of patients. GBV-C/HGV is probably "harmless" to its host, due to a long history of mutual adaptation. Following the discovery of the GBV-C/HGV, the genome of a novel DNA virus, designated as the TT virus (TTV), was discovered

Table 5. Genotypic distribution of GBV-C/HGV in different countries, as determined by PCR using the genotype-specific primers

| Country | *n* | Genotype | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 | 2+3 | 3+4 | 2+4 | UC* |
| Ghana | 22 | 22 (100) | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| U.S. | 14 | 2 (14) | 11 (79) | 1 (7) | 0 | 0 | 0 | 0 | 0 |
| Spain | 16 | 0 | 16 (100) | 0 | 0 | 0 | 0 | 0 | 0 |
| Egypt | 39 | 3 (8) | 36 (92) | 0 | 0 | 0 | 0 | 0 | 0 |
| Nepal | 7 | 0 | 4 (57) | 3 (43) | 0 | 0 | 0 | 0 | 0 |
| Thailand | 3 | 0 | 3 (100) | 0 | 0 | 0 | 0 | 0 | 0 |
| Myanmar | 47 | 0 | 27 (58) | 5 (11) | 11 (23) | 2 (4) | 0 | 0 | 2 (4) |
| Vietnam | 28 | 0 | 5 (18) | 4 (14) | 17 (60) | 1 (4) | 0 | 0 | 1 (4) |
| Cambodia | 4 | 0 | 3 (75) | 0 | 1 (25) | 0 | 0 | 0 | 0 |
| Japan | 20 | 1 (5) | 1 (5) | 14 (70) | 0 | 2 (10) | 1 (5) | 1 (5) | 0 |
| Philippines | 2 | 0 | 0 | 2 (100) | 0 | 0 | 0 | 0 | 0 |
| Bolivia | 49 | 0 | 18 (37) | 26 (53) | 0 | 4 (8) | 0 | 0 | 1 (2) |
| Total | 251 | 28 (11) | 124 (49) | 55 (22) | 29 (12) | 9 (4) | 1 (0.4) | 1 (0.4) | 4 (1.6) |

Number in parenthesis indicate percentages. *Unclassified (ref. 42)

recently from a patient with post-transfusion non-B, non-C acute hepatitis by representational difference analysis (45). It appears to be a ubiquitous virus. The TTV infection does not appear to have a significant effect on liver disease and therefore it also could be considered as an "orphan virus". However, continued study of these novel viruses in non-liver-related clinical settings will help to determine whether the virus contributes to other human diseases with unknown etiology.

## ACKNOWLEDGMENTS

I thank Dr. Takeshi Kurata (Deputy Director-General) and Dr. Tetsutaro Sata (Director of Pathology) of the National Institute of Infectious Diseases for their continuous encouragement during this study. Drs. Hideo Naito, Tamiko Saito, Nami Konomi, Tomoko Inami, Naoto Aiba, Shigeki Hayashi, Yutaka Takebe, Koichi Ishikawa, Khim Maung Win, Vo Xuan Ouang, and David Y. Zhang are to be thanked for their kind collaboration.

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