Serological evidence of hepatitis E virus infection in an indigenous North American population

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BACKGROUND: Hepatitis E virus (HEV) infections are thought to be uncommon in North America. Recently, HEV transmission has been reported following the consumption of deer meat. Because deer are closely related to caribou and caribou meat is a staple of the Canadian Inuit and the American Eskimo diet, the present study explored the seroprevalence of HEV infection in an isolated Canadian Inuit community.

METHODS: Stored sera were thawed and tested for immunoglobulin (Ig) G and IgM anti-HEV by ELISA, and tested for HEV-RNA by reverse transcriptase polymerase chain reaction.

RESULTS: The study consisted of 393 sera (representing approximately 50% of the community’s inhabitants). Eleven samples (3%) were IgG anti-HEV-positive. Their mean age was 29±8 years and three were male. Two of 11 (18%) were also IgM anti-HEV-positive. All IgG anti-HEV-positive individuals were HEV-RNA-negative. Liver biochemistry was normal in all. Seven of 11 (64%) were also positive for anti-hepatitis A virus, five (46%) were hepatitis B virus seropositive and none (0%) were positive for anti-hepatitis C virus. There were no associations between infections with HEV and other hepatotropic viruses. Serological testing was negative for HEV infection in 25 caribou from an adjacent region.

CONCLUSION: The results of the present study showed that serological evidence of HEV infection was present in 3% of the observed Canadian Inuit population; the presence of IgM anti-HEV suggested recent infection and HEV did not appear to coinfect with other common hepatotropic viruses. The source of HEV infection in the population remains unclear. These findings are interesting but preliminary. Additional data are required to determine whether HEV infections are responsible for otherwise unexplained acute hepatitis in the Canadian Inuit population and visitors returning from northern North American communities.

Key Words: Acute hepatitis; Canada; Hepatitis E virus; Liver; Liver disease; Viral hepatitis

Thus, to diagnose acute HEV, either immunoglobulin (Ig) M anti-HEV or RT-PCR testing can be used (7,8). For evidence of previous infection, IgG anti-HEV is considered to be highly sensitive and specific (greater than 95%, respectively) (9,10).

HEV is transmitted by the fecal-oral route (1). Typically, this involves ingesting water or food contaminated by infected feces (6). Recent studies (11-14) have reported that HEV can be detected in primates, swine and deer, and that humans can become infected by consuming the meat of these animals. Because North American caribou are closely related to deer, it is possible...
that caribou may also carry the HEV infection. This could repre-
sent a potential health risk to the Canadian Inuit because caribou
meat is a staple of their diet. The purpose of the present study was
to document the prevalence of serological markers for HEV infec-
tion in a remote, northern Canadian Inuit community.

PATIENTS AND METHODS

Study population and serological testing

The study community is an inland settlement south of the Arctic
Circle and north of the Canadian treeline. The size of the com-
munity at the time the samples were collected (1980) was approxi-
mately 800 individuals (15).

Most families within the community derive their income from
hunting, fishing or federal government assistance. Travel in and
out of the community is largely by plane, and is limited to indi-
viduals visiting family members in adjacent communities and
medical evacuations to the nearest medical centre in Churchill,
Manitoba. Most, if not all, community inhabitants have not trav-
elled outside Canada. Tourist activity is limited.

Caribou meat, fish and processed food from southern Canadian
centres constitute the staple diet. Caribou meat is usually eaten
raw when obtained by hunting ‘on the land’. Caribou meat stored
outside of dwellings within the community is either eaten raw,
fried, or cooked in warm or hot water for 30 min to 45 min.

Human serum samples from inhabitants of the community,
which were collected in 1980 and kept frozen at –80°C, were
thawed for testing (15). For each individual, their dwelling within
the community was assigned a number and their status within the
community was recorded. Caribou individuals visiting family members in adjacent communities and
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Manitoba. Most, if not all, community inhabitants have not trav-
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Human serum samples from inhabitants of the community,
which were collected in 1980 and kept frozen at –80°C, were
thawed for testing (15). For each individual, their dwelling within
the community was assigned a number and their status within the
family unit (eg, father, mother, son, etc) was recorded. Caribou
sera (kindly provided by Drs Rick Farnell, Phillip Merchant and
Dorothy Cooley, Yukon Department of Environment,
Whitehorse, Yukon) were derived from the Porcupine and
Chisana Woodland herds in regions adjacent to the study com-
nunity. Serological testing was performed using commercially
available kits from Genelabs Diagnostics (USA). ELISA was used
to detect IgG and IgM anti-HEV following the manufacturer’s
instructions. Samples that were IgG anti-HEV-positive were
retested in duplicate. If all three test results were positive, the
sample was considered positive and was further tested (in duplicate)
for IgM anti-HEV and HEV RNA. HEV RNA testing was
performed by RT-PCR. Serum RNA was isolated using a High
Pure Viral RNA Kit (Roche Diagnostics, Canada) and RT-PCR
was performed as described by Mizuo et al (16). Positive controls
were provided by Dr C Osiowy at the National Microbiology
Laboratory in Winnipeg, Manitoba. Serological testing for hepati-
sis A virus (HAV), hepatitis B virus (HBV) and hepatitis C virus
(HCV) infections was performed as described previously (15,17).

All individuals or their guardians provided informed consent
in 1980 for future testing of newly discovered hepatotropic viruses
and/or variants of established causes of viral hepatitis. The study
protocol was approved by the University of Manitoba’s Biomedical
Research Ethics Board.

Statistics

Categorical variables were evaluated using the $\chi^2$ analysis. The $\chi^2$
test of association (or F test when warranted) was used to examine
whether the presence of anti-HEV was associated with positive
results of serological testing for HAV, HBV or HCV. Continuous
variables were compared using Student’s t test. Statistical signifi-
cance was achieved when P values fell below 5% in all analyses.
Statistical analyses were performed using NCSS and PASS statis-
tical software (NCSS, USA).

RESULTS

The study sample consisted of 393 sera (representing approxi-
mately 50% of the community inhabitants), 11 (3%) of whom
were IgG anti-HEV-positive (Table 1). Their mean age was
29±8 years and three of the 10 (30%) in whom sex was known
were male. Two of the 11 samples (18%) were IgM anti-HEV-
positive. Neither of these nor the remaining nine IgG anti-
HEV-positive samples were HEV-RNA-positive. All
11 anti-HEV-positive individuals had normal liver enzyme tests
( serum aminotransferases, alkaline phosphatase and gamma
glutamyl transpeptidase levels) and were rheumatoid factor-
negative. Seven (64%) were anti-HAV-positive, five (46%)
were HBV seropositive (either HBV surface antigen, or anti-
body to HBV surface or core antigen-positive) and none (0%)
were anti-HCV-positive. There were no associations between
the presence of anti-HEV and positive serology for HAV, HBV
or HCV. When analyzed with respect to family dwellings, none

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<th>IgM anti-HEV</th>
<th>HEV RNA</th>
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<th>Anti-HCV</th>
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</table>

*Mean ± SD. – Negative; + Positive; HAV Hepatitis A virus; HBV Hepatitis B virus-seropositive (either HBV surface antigen or antibody to HBV core or surface antigen-positive); HCV Hepatitis C virus; IgG/M Immunoglobulin G/M; NT Not tested
of the 11 anti-HEV-positive individuals resided in the same dwelling, nor were they first-degree relatives to each other.

The age and sex distributions of the remaining 382 anti-HEV-negative individuals are also provided in Table 1. Their mean age was 22±1 years and 194 (51%) were male. Two hundred seven (68%) had been previously documented to be anti-HAV-positive, 86 (22%) HBV seropositive and four (1.0%) anti-HCV-positive (15,17).

IgG and IgM anti-HEV testing in all 25 caribou sera was negative.

DISCUSSION

To date, essentially all reported cases of HEV in Canada have been described in travellers returning from tropical and/or developing nations (18,19). The prevalence of HEV in a community-based North American population or, indeed, any community-based population throughout the world has yet to be reported. Although the results of the present study are limited to one community of a selected Canadian population, they do suggest that HEV infections unrelated to travel or working with pigs are occurring in North America and specifically within this northern indigenous population.

The 3% prevalence rate described in the present study should only be considered to be an estimate of the true prevalence of HEV in this region. The half-life of IgG anti-HEV is relatively short and may become negative within one year of infection (20,21). Thus, other community inhabitants may have been infected with HEV in the more remote past, but were seronegative at the time blood samples were obtained. False-positive results must also be considered. Although all positive specimens were tested in triplicate and were negative for rheumatoid factor, the presence of a not yet recognized cross-reacting antibody cannot be excluded.

Two of the 11 anti-HEV-positive samples also tested positive for IgM anti-HEV, consistent with recent HEV infections. However, the normal liver enzymes and negative HEV-RNA testing in these samples would be unusual for acute HEV. Whether these specimens are false-positive for IgM anti-HEV or reflect the known decline in serum aminotransferases and viral RNA stability with storage of sera over prolonged periods of time remains to be determined (22,23).

CONCLUSION

Approximately 3% of this Canadian Inuit population have serological evidence of HEV infection. These results support the need for further studies to determine whether HEV is the cause of unexplained acute hepatitis in individuals residing in or visiting the northern regions of North America.

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REFERENCES


