Host risk factors and autochthonous hepatitis E infection

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Introduction **In developed countries autochthonous hepatitis E infection is caused by hepatitis E virus (HEV) genotype 3 or 4 and mainly affects middle aged/elderly men. Host factors might explain why older men develop clinically overt disease.**

Methods **Retrospective review of 53 patients with symptomatic autochthonous hepatitis E infection to determine putative host risk factors. Patients were compared with 564 controls with adjustment for age and sex. Anti-HEV seroprevalence was determined in controls and 189 patients with chronic liver disease.**

Results **Mean age of the patients was 62.4 years, 73.6% were men. Compared with controls, patients with hepatitis E were more likely to drink at least 22 U alcohol/week (OR = 9.4; 95% confidence interval = 3.8–25.0; $P < 0.001$). The seroprevalence of anti-HEV IgG in controls increased with age ($P < 0.001$) but was similar in men and women. There was no association between alcohol consumption and anti-HEV IgG seroprevalence in the control group. There was no difference in the anti-HEV IgG seroprevalence between the controls and patients with chronic liver disease of all aetiologies, but seroprevalence was higher in controls (13.8%) than patients with alcoholic liver disease (4.8%, $P = 0.04$).**

Conclusion **Clinically apparent hepatitis E infection is more common in individuals who consume at least 22 U alcohol/week. Patients with established chronic alcoholic liver disease have a low seroprevalence compared with controls. The reason for this observation is uncertain, but patients with alcoholic liver disease have clinically severe disease with a high mortality when exposed to HEV. The low seroprevalence in this group may represent a 'culled' population.** ***Eur J Gastroenterol Hepatol*** **23:1200–1205** © **2011 Wolters Kluwer Health | Lippincott Williams & Wilkins.**

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Introduction

Autochthonous (locally acquired) hepatitis E virus (HEV) infection is an emerging health issue in developed countries [1]. It has a predilection for older men and generally causes a self-limiting acute hepatitis. The overall mortality rate is 5–10% [2–4], but the prognosis is poor in patients with underlying chronic liver disease, with case fatality rates of up to 70% [5–7].

Recently, chronic HEV infection with rapidly progressive cirrhosis has been demonstrated in immunosuppressed transplant recipients [8], individuals with haematological malignancies [9] and patients with HIV infection [10]. Autochthonous hepatitis E infection in developed countries is mainly caused by genotype 3 HEV (and genotype 4 HEV in China and Japan) and may be a porcine zoonosis. Cases of hepatitis E infection have been documented after the consumption of uncooked or poorly cooked pork meat [11], but in most cases the exact route of infection remains uncertain.

In developed countries, in contrast to endemic areas in the developing world, clinical cases of autochthonous hepatitis E infection have been documented predominantly in the middle aged and elderly and more commonly in men [12–15]. The reason for this observation is not understood. It seems unlikely that this can be explained by increased exposure to HEV in these groups, as HEV acquisition has been shown to be independent of age, at least in the UK [16]. Hepatitis E infection results in a wide range of clinical disease expression from cases that are asymptomatic to severe hepatitis with hepatic failure [2–4]. It is possible that host factors might explain why middle aged/elderly men develop clinically overt disease and present to medical attention.

To date, little is known with regard to host factors and clinical disease expression in hepatitis E infection. The aim of this study was to investigate the role of host factors in hepatitis E infection using a case–control approach.

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Methods

Case definitions

Autochthonous hepatitis E infection was defined as:

(1) biochemical evidence of hepatitis (raised serum transaminase);
(2) no recent (< 3 months) travel to an area considered 'endemic' for hepatitis E;
(3) anti-HEV IgM positive in serum; and/or
(4) rising serum anti-HEV IgG; and/or
(5) HEV RNA detected in the serum by PCR.

Symptomatic autochthonous hepatitis E infection was defined as:

(1) Autochthonous hepatitis E infection (see above) with temporally-related symptoms caused by the infection [4].

Patients

Over the last 10 years, 56 patients with autochthonous hepatitis E infection have been documented in Cornwall and Devon, UK. Of these 56 patients, three patients had no symptoms and 53 had symptoms. The clinical and laboratory features of most of these cases have previously been described elsewhere [4]. None of the patients had travelled to an area endemic for HEV. Thirty-six of the patients were HEV PCR positive, all HEV genotype 3. Fifty-three patients with symptomatic HEV infection were retrospectively reviewed to determine age, sex, past medical history, drug history and dietary habits (including the quantity of alcohol consumed and whether the patient was vegetarian). These data were analysed with descriptive statistics to see if any putative host risk factors emerged (Table 1).

Table 1 Host factors in 53 patients with symptomatic autochthonous hepatitis E

Mean age (range)	62.3 (38–83 years)
Past medical history	
Hypercholesterolaemia	$n=21$
Diabetes	$n=11$
Hypertension	$n=11$
No past medical history	$n=10$
Ischaemic heart disease	$n=9$
Chronic renal disease	$n=5$
Chronic respiratory disease	$n=4$
Chronic liver disease	$n=3$
Peripheral vascular disease	$n=3$
Venous thromboembolism	$n=3$
Miscellaneous[a]	$n=32$
Prescription drugs taken at onset of HEV infection	
Statin	$n=11$
Oral hypoglycaemics/insulin	$n=10$
ACE inhibitors	$n=10$
No drug therapy	$n=8$
Aspirin	$n=6$
Antihypertensives	$n=6$
Steroid/β-2 agonist inhalers	$n=6$
Proton pump inhibitors	$n=5$
Prednisolone	$n=3$
NSAIDs	$n=3$
Miscellaneous[b]	$n=34$
Weekly alcohol consumption (U/week)[c] at onset of HEV infection	
Nil	$n=9$
1–3	$n=4$
4–21	$n=15$
≥ 22	$n=21$
Unknown	$n=4$
Vegetarian diet	0/53

ACE, angiotensin-coverting enzyme; HEV, hepatitis E virus.
[a]Miscellaneous included ($n=1$ unless stated): cerebrovascular accident ($n=2$), polymyalgia rheumatica ($n=2$), osteoarthritis ($n=2$), peripheral neuropathy ($n=2$), hypothyroidism ($n=2$), peptic ulcer disease treated by surgery ($n=2$), reflux disease, atrial fibrillation ($n=2$), prostate cancer, bladder cancer, Crohn's colitis, HIV, hysterectomy, herpes zoster, steroid myopathy, psoriasis, macular degeneration, myelodyplasia, Whipple's procedure, epilepsy, benign prostatic hypertrophy, dementia, aortic stenosis, ischaemic colitis, renal stones.
[b]Miscellaneous included ($n=1$ unless stated): benzodiazepines ($n=2$), thyroxine ($n=3$), clopidogrel ($n=2$), glucosmaine, hormone replacement therapy ($n=2$), finasteride ($n=2$), calcichew ($n=2$), paracetamol, digoxin, carbemazepine, mesalazine, methotrexate, movicol, diltaizem, motelukast, isosorbide mononitrate, quinine sulphate, zolodex, fexofanadine, cephalexin, antiretroviral therapy, uniphylin, monomax, alendronate, warfarin ($n=2$), zafirlukast.
[c]1 unit = 10 ml of pure alcohol.

Controls

After providing informed consent, 564 patients were used as controls. These individuals were aged at least 18 years with no history of liver disease and were identified from in patients and out patients attending the Royal Cornwall Hospital, Truro and a single General Practice in Cornwall. Details of their alcohol consumption, diet, history of hypercholesterolaemia, diabetes and drug therapy were recorded prospectively. A blood sample was taken for anti-HEV IgG testing.

Patients with chronic liver disease

One hundred and eighty-nine patients with clinical and/or radiological and/or histological evidence of chronic liver disease were identified at the Royal Cornwall Hospital, Truro. After informed consent, the aetiology of their chronic liver disease was recorded and a blood sample taken for anti-HEV IgG testing.

Hepatitis E virus serology

Serum samples from the controls and patients with chronic liver disease were tested for anti-HEV IgG antibodies using the Wantai HEV IgG EIA immunoassay (Wantai Biological Pharmacy Enterprise, Beijing, China). This assay uses antigens encoded by a structural region of ORF-2 from a Chinese isolate of genotype 1 HEV [17]. The assay was performed according to the manufacturer's instructions. Sera giving an absorbance greater than the cut-off value were considered to be positive for anti-HEV IgG.

Statistics

Associations between hepatitis E infection and host risk factors were assessed using logistic regression models after adjustment for age and sex. Exposure effects were expressed as odds ratios (ORs) with 95% confidence

intervals. The shape of the relationship between age and anti-HEV IgG seroprevalence amongst the controls was explored using logistic regression models with linear and quadratic terms for age. Differential effects of age by sex were assessed through the inclusion of appropriate interaction terms in the models. Age–sex adjusted logistic regression models were also used to compare anti-HEV IgG seroprevalence in cases of chronic liver disease and controls. All statistical analyses were carried out using the R software [18].

This study had ethical approval from the Plymouth and Cornwall Ethics Committee.

Results

Fifty-three patients with hepatitis E infection were identified; their host risk factors are shown in Table 1. Cases of hepatitis E were mainly observed in older men (Fig. 1). The mean age of the patients was 62.4 years (median 63 years, range 36–83 years) and 39 (73.6%) were men. Forty-five of 53 (84.9%) cases occurred in patients aged 40–80 years, with a peak in the sixth and seventh decades. Twenty-one (39.6%) of the 53 patients had previously documented hypercholesterolaemia, 21 (39.6%) consumed at least 22 U alcohol/week, 11 (20.8%) were diabetic, 11 (20.8%) were taking a statin. None of the patients were vegetarian, and all patients ate pork or pork containing products.

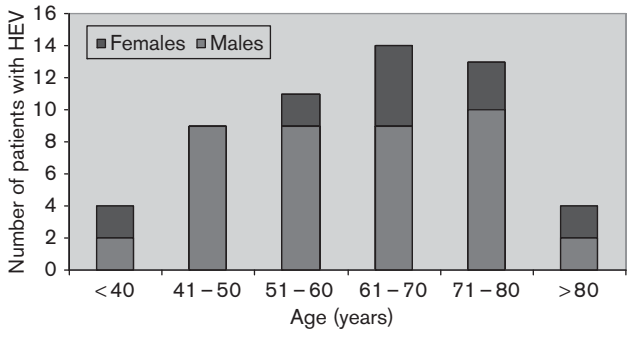
The demographic profile of the control group is shown in Table 2 (39.2% men; mean and median ages 52.6 and 53 years, respectively). There were significant differences in age and sex between patients and controls, with more women and younger individuals in the control group. After adjusting for age and sex, compared with controls, patients with hepatitis E were significantly more likely to drink at least 22 U alcohol/week (OR = 9.4 compared with nondrinkers; 95% confidence interval = 3.8–25.0; $P < 0.001$). Table 3 shows the association between alcohol consumption and hepatitis E infection was present in both men and women, as well as amongst younger (age ≤ 60) and older patients (age > 60). Twenty-one percent of patients were diabetic, compared with 8.2% of controls: this difference just failed to reach statistical significance ($P = 0.07$). There was no association between hypercholesterolaemia or use of a statin and anti-HEV IgG positivity. None of the patients were vegetarian compared with 4.1% of controls: it was not possible to calculate an OR for these data (Table 2).

Table 2 Hepatitis E virus host factors: a comparison of symptomatic hepatitis E virus patients ($n = 53$) and controls ($n = 564$)

	N	Controls (%)	HEV patients (%)	OR	95% CI	*P* value
Sex						
Males	260	39.2	73.6	1.00		
Females	357	60.8	26.4	0.23	0.12–0.43	<0.001
Age (years)						
<40	148	25.6	7.5	1.00		
40–50	118	19.7	13.2	1.95	0.57–7.69	0.30
50–60	125	20.2	20.8	2.97	0.97–11.05	0.07
60–70	116	18.1	26.4	4.38	1.50–15.98	0.01
70+	109	16.3	32.1	6.60	2.32–23.78	0.00
Diabetes						
No	560	91.8	79.2	1.00		
Yes	57	8.2	20.8	2.04	0.9–4.35	0.07
Type I	8	1.2	1.9			
Type II	49	6.9	18.9			
Hypercholesterolaemia						
No	292	47.7	43.4	1.00		
Yes	181	28.2	41.5	1.35	0.71–2.58	0.35
Missing	144	24.1	15.1	1.76	0.65–4.48	0.24
Statin						
No	509	85.5	50.9	1.00		
Yes	92	14.4	20.8	1.45	0.62–3.23	0.37
Missing	16	0.2	28.3	360.57	58.0–7311	<0.001
Alcohol (U/week)						
0	221	37.4	18.9	1.00		
1–3	97	16.5	7.5	0.86	0.23–2.73	0.81
4–21	240	39.9	28.3	1.37	0.59–3.32	0.47
22+	54	6.0	37.7	9.42	3.78–24.95	<0.001
Missing	5	0.2	7.5	70.25	7.05–1784	0.0013
Vegetarian						
No	594	96.00	100	1.00		
Yes	23	4.10	0.00	NA	NA	0.99

CI, confidence interval; HEV, hepatitis E virus; NA, not available; OR, odds ratio.

Fig. 1



Age and sex distribution of 53 patients with symptomatic autochthonous hepatitis E infection in Cornwall and Devon.

The seroprevalence of anti-HEV IgG in the control group increased with age ($P < 0.001$; Fig. 2). The observed difference in anti-HEV IgG seroprevalence between men and women was not statistically significant ($P = 0.5$). There was no association between alcohol consumption and anti-HEV IgG seroprevalence in the control group (before and after adjusting for age and sex, $P = 0.3$ and $P = 0.2$ respectively).

Of the 189 patients with chronic liver disease, 104 (55%) were men and 85 (45%) were women. The mean age was 56.8 years (median age 58 years, range 22–91 years). The aetiologies of the chronic liver disease were: alcohol $n = 64$ (33.9%); nonalcoholic fatty liver disease $n = 39$ (20.6%); autoimmune hepatitis $n = 28$ (14.8%); chronic hepatitis C infection $n = 25$ (13.2%); primary biliary cirrhosis $n = 18$ (9.5%); miscellaneous $n = 17$ (9%). The latter group included patients with cryptogenic cirrhosis ($n = 4$), chronic hepatitis B infection ($n = 3$), primary

Table 3 The association between alcohol consumption and hepatitis E virus infection stratified by age and sex

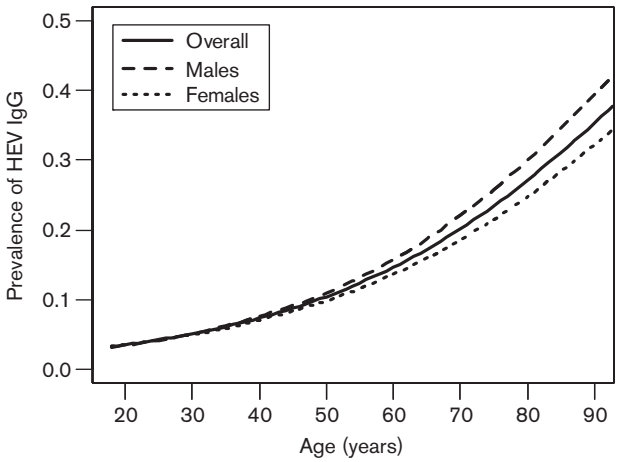
	HEV controls		HEV patients				
	Alcohol consumption		Alcohol consumption				
	<22 units	≥ 22 units	<22 units	≥ 22 units	OR[a]	95% CI	*P* value
Sex							
Males	193	28	18	17	6.51	3.01–14.09	<0.001
Females	336	6	11	3	15.27	3.37–69.17	<0.001
Age							
≤ 60 years	343	25	11	10	12.47	4.83–32.18	<0.001
>60 years	185	9	18	10	11.42	4.11–31.74	<0.001

CI, confidence interval; HEV, hepatitis E virus; OR, odds ratio.
[a]Ratio of odds of HEV infection given high alcohol consumption to odds of HEV infection given low alcohol consumption.

sclerosing cholangitis ($n = 2$), haemochromatosis with cirrhosis ($n = 2$), 'nonspecific hepatitis' with fibrosis ($n = 2$), secondary biliary cirrhosis ($n = 1$), not determined ($n = 1$).

Table 4 shows the distributions of anti-HEV IgG seroprevalence by age and sex for patients with chronic liver disease and controls. After adjusting for age and sex, comparison of the control group to the patients with established liver disease of all aetiologies ($n = 189$) showed no difference in the anti-HEV IgG seroprevalence between these populations ($P = 0.8$). However, there was a significantly higher anti-HEV IgG seroprevalence amongst the controls (64/464, 13.8%) than patients with alcoholic liver disease (3/62, 4.8%) after accounting for age and sex ($P = 0.04$). In contrast, after adjusting for age and sex, there was no difference in anti-HEV IgG seroprevalence between controls and patients with chronic liver disease of a nonalcoholic aetiology ($P = 0.3$).

Fig. 2



Estimated seroprevalence of anti-hepatitis E virus (HEV) IgG amongst the control population as a function of age. The solid line shows age-specific prevalence estimates calculated from a logistic regression model fitted to the full control sample. The model was specified with the log-odds of anti-HEV IgG seroprevalence expressed as an additive linear function of age and sex. The log-odds were converted to a prevalence estimate using an inverse logit transformation. The dashed lines show prevalence estimates for men and women separately as a function of age, calculated from sex-specific logistic regression models.

Discussion

In developed countries HEV causes a spectrum of illness from asymptomatic infection to fulminant hepatic failure [4]. Seroprevalence rates of 10–30% in some US states [19,20] and some EU countries [21,22], suggest that asymptomatic infection is common as overt infection is uncommon in these areas. A recent study of an outbreak of HEV genotype 3 infection on a cruise ship found that 67% of patients had no symptoms related to hepatitis E [23]. Previous studies have shown that clinically overt cases of locally acquired hepatitis E in developed countries are more common in middle aged and elderly men [12–15]. This study confirms these findings. The reason for this observation is uncertain. It could be that middle aged and elderly men have increased exposure to HEV or that host factors may make these patients more likely to develop clinically overt disease if exposed.

Though overt hepatitis E is common in men, this study indicates that there is no difference in anti-HEV IgG seroprevalence between men and women in the control group. Previous studies also have shown little evidence to suggest that middle aged/elderly men have increased rates of infection with HEV [16]. Data from France and Spain have shown no sex difference in anti-HEV seroprevalence [22,24] and a UK study showed that elderly men had only a modestly (OR 1.6) increased seroprevalence compared with elderly women [4]. Most previous studies show that anti-HEV seroprevalence increases with age [16,21,22,24]. However, anti-HEV IgG antibodies have been documented in people of all ages, and it seems that the rate of HEV acquisition is independent of age [16]. The high seroprevalence rates observed in the elderly men in the UK is thought to be because of a combination of cumulative exposure over time and a 'cohort' effect, reflecting a relative higher burden of exposure to HEV in previous decades [16].

It seems from this study that alcohol consumption is an important risk factor in the clinical expression of HEV infection. Consumption of at least 22 U alcohol/week

Table 4 Antihepatitis E virus immunoglobulin G seroprevalence for patients with chronic liver disease ($n=189$) and controls ($n=464$) by age and sex

	Controls			Patients with chronic liver disease		
	Anti-HEV IgG			Anti-HEV IgG		
	Negative	Positive	Seroprevalence (%)	Negative	Positive	Seroprevalence (%)
Sex						
Males	161	28	13.1	92	12	15.5
Females	239	36	14.8	71	13	11.5
Age (years)						
<40	105	6	5.4	18	1	5.3
40–50	64	6	8.6	37	4	9.8
50–60	81	12	12.9	42	5	10.6
60–70	79	19	19.4	48	7	12.7
70+	71	21	22.8	18	8	30.8

HEV, hepatitis E virus; IgG, immunoglobulin G.

(the current UK Department of Health's recommended maximum weekly intake for men) is strongly associated with acute hepatitis E in our patient population. This accords with analysis of a HEV genotype 3 outbreak which identified alcohol consumption as a key risk factor [23]. We do not believe this association is because of increased exposure to HEV in drinkers, as there was no association between alcohol consumption and HEV seroprevalence in the control individuals. It is more likely that heavy alcohol consumption might produce subclinical liver injury (steatosis and/or fibrosis) which makes symptomatic hepatitis E more likely after exposure to the virus.

Our data shows that there is no difference in HEV acquisition between patients with chronic liver disease and control individuals. However, patients with established chronic alcoholic liver disease have a significantly lower anti-HEV seroprevalence than control individuals. This is an intriguing observation, and its explanation is uncertain. HEV infection in the context of chronic liver disease carries a poor prognosis, with mortality rates approaching 70% [5–7]. If patients with alcoholic cirrhosis are prone to developing severe hepatitis (with its associated high mortality) after exposure to HEV, the very low anti-HEV seroprevalence rates observed in these patients may reflect a 'culled' population, where the majority of those who acquired HEV have died.

Over 20% patients with the symptomatic hepatitis E in this study were in people with diabetes. Compared with the control group, 8.2% of whom were people with diabetes, this just failed to reach statistical significance. A previous study in England showed that four of 13 patients with autochthonous hepatitis E infection were people with diabetes [25]. A larger number of patients is required to determine whether diabetes is a risk factor for overt infection with HEV. The main limitation of this study is the way in which the control individuals were selected. Ideally, such individuals would have been randomly selected from the general population. Instead, our controls were drawn from patients accessing medical care. It is likely that selecting the control group in this way increased the number of diabetics and heavy alcohol consumers in the control group compared with the normal population in the community, as such individuals tend to be heavier users of primary and secondary healthcare services. This may have led us to underestimate the contribution of these factors to the risk of developing acute hepatitis E.

In conclusion, clinically apparent autochthonous hepatitis E infection is more common in older men and in individuals who consume at least 22 U alcohol/week. This is unlikely to be because of increased exposure to HEV in heavy alcohol consumers as their seroprevalence did not differ from more moderate drinkers. Patients with established chronic liver disease because of alcohol have a low seroprevalence compared with controls. The reason for this observation is uncertain, but it may be that patients with alcoholic liver disease have clinically severe disease with a high mortality when exposed to HEV.

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Conflicts of interest

Dr Dalton had travel and accommodation costs reimbursed and consultancy fees paid by GlaxoSmithKeline and Wantia Biological enterprise.

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