HEV: inquadramento epidemiologico-clinico

Prof.ssa Caterina Sagnelli

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HEV

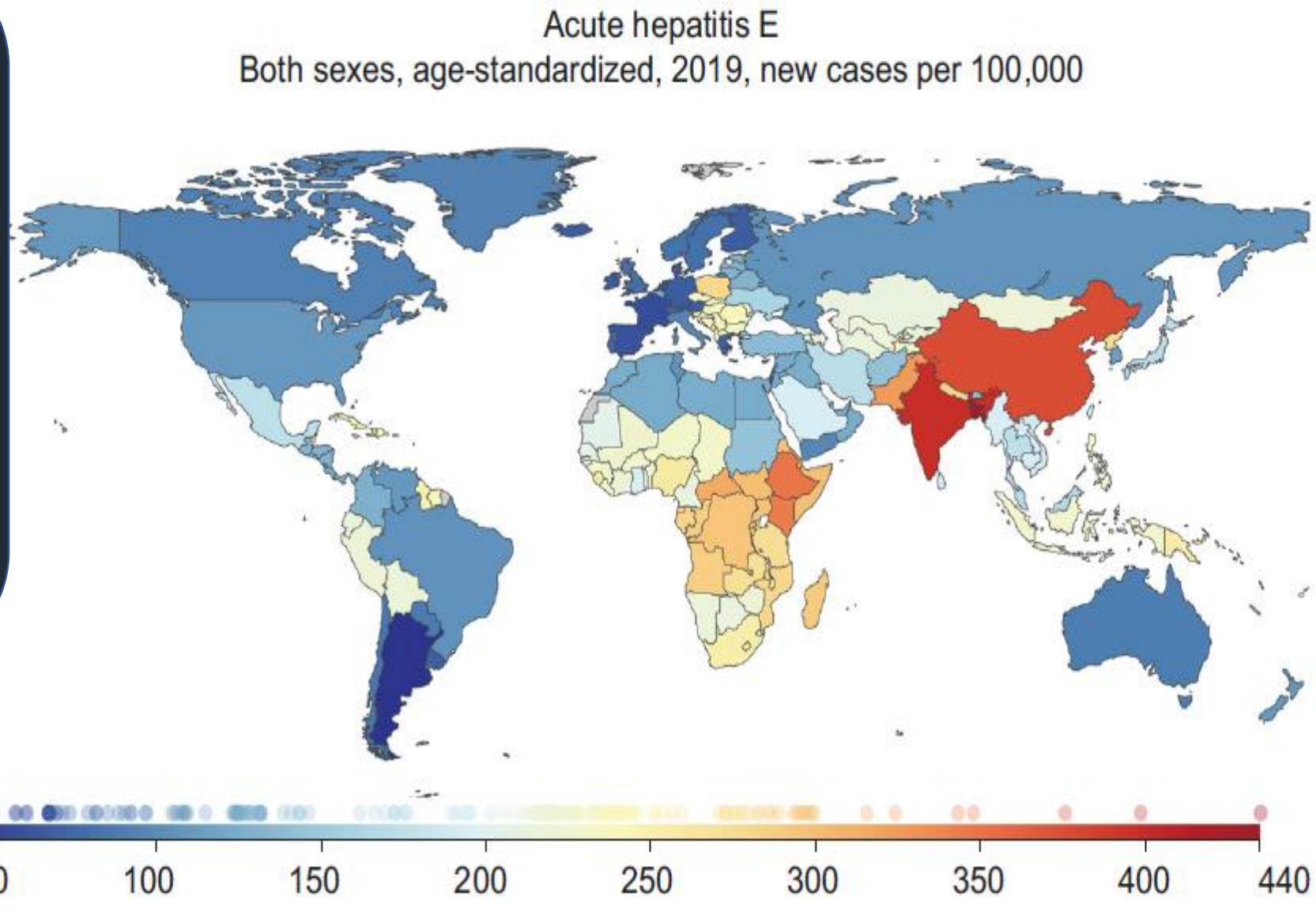
- **HEV: the first time in 1983 in stool samples.**
- **HEV is common in low-middle-income countries with limited access to water, sanitation, hygiene and health services.**
- **The outbreaks follow periods of faecal contamination of drinking water supplies and may affect several hundred to several thousand persons.**
- **Outbreaks: in areas of conflict and humanitarian emergencies.**

Nel 1955-1956, primo focolaio di HEV in New Delhi, India (water), 29300 individui

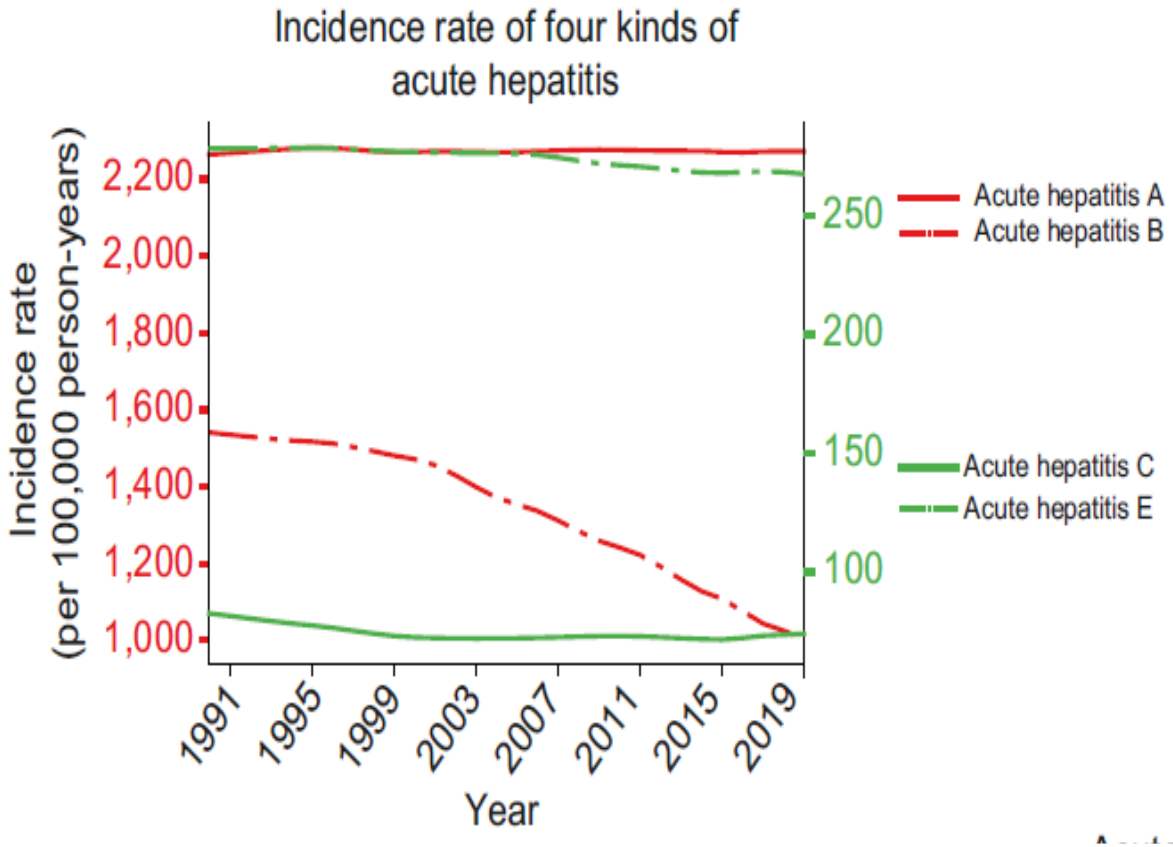
Year(s)	Country	Mode of Transmission	Reported
2013	Cameroon	Waterborne	33
2014–2015	Bangladesh	Waterborne	103
2014–2016	India	Waterborne	17
2016–2017	Chad	Waterborne	1293
2017–2018	Nigeria	Contamination of drinking water	1376
2014–2017	Bangladesh	Waterborne	661
2018	Central African Republic	Waterborne	149
2018	South Sudan	Waterborne	161
2019	Pakistan	Waterborne	300
2017–2020	Namibia	Waterborne	7247

The Global Epidemiological of HEV

HEV1,2 associati a **20,1 milioni di nuove infezioni** all'anno in Asia e Africa con **3,4 milioni** di HEV sintomatica, **70000 decessi per** insufficienza epatica acuta
e **3000 nati morti**



Burden of acute HEV



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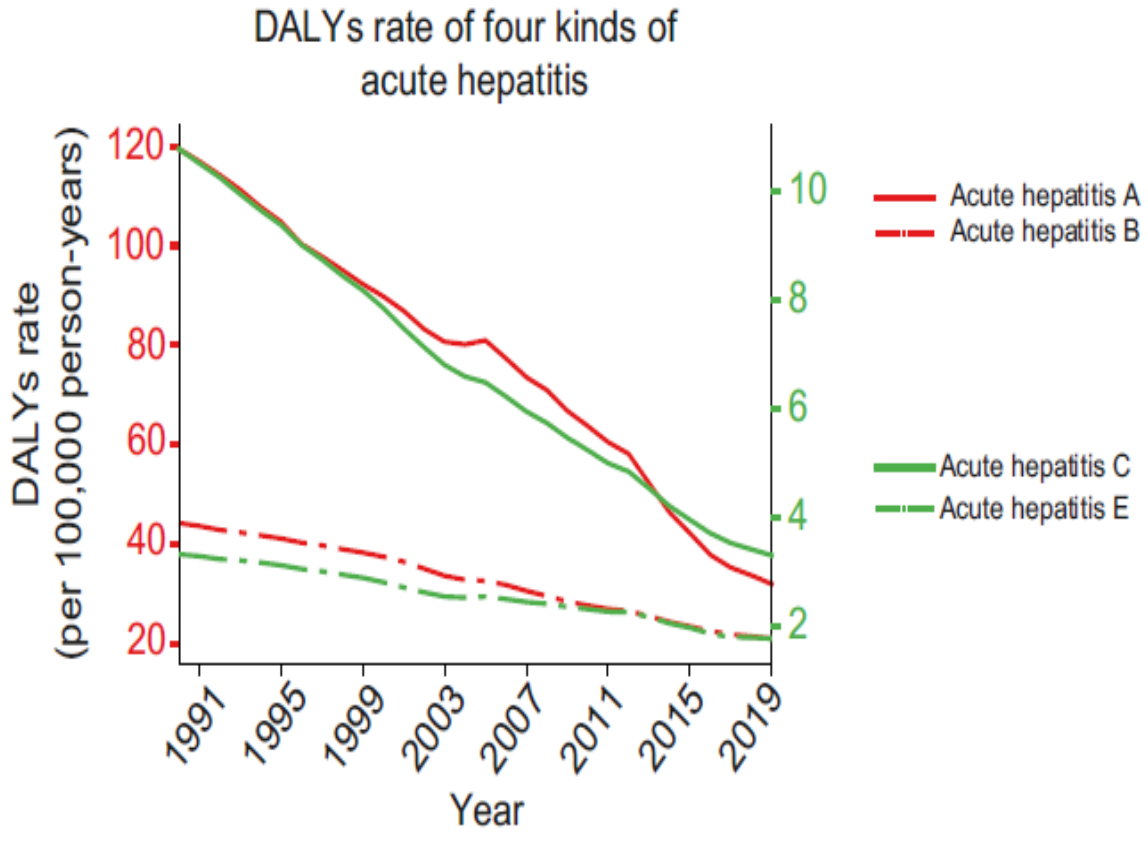


East and South Asian regions shoulder heaviest burden of AHE.

highest incidence rates: Bangladesh (435.3 [353.6–518.4] per 100,000 p-y), followed India, China. highest DALY rates: Tonga (8.1 [4.4–13.4] per 100,000 p-y), followed Zimbabwe, India. the lowest age-standardized incidence rate: Argentina, San Marino, France. lowest DALY rates: Austria, Switzerland, Israel.

dominant genotype in China has shifted from HEV1 to HEV4, reflect improvement in sanitary

In Europa, la prevalenza delle infezioni da HEV varia a seconda delle regioni

- HEV3 è iperendemico nel sud-ovest della Francia: tasso prevalenza > 50%.
- È endemico nel nord della Francia, Belgio, Paesi Bassi e Germania. I tassi di prevalenza fino al 30%.
- HEV è l'epatite virale acuta 2013-2015 nei Paesi Bassi.
- In 30 paesi europei, il numero di casi di infezione da HEV è passato da 514 all'anno nel 2005 a 5617 nel 2015.
- Il tasso di prevalenza degli Ab anti-HEV in Asia e Africa è del 10-40% con frequenza crescente nei gruppi di età > 50 anni.

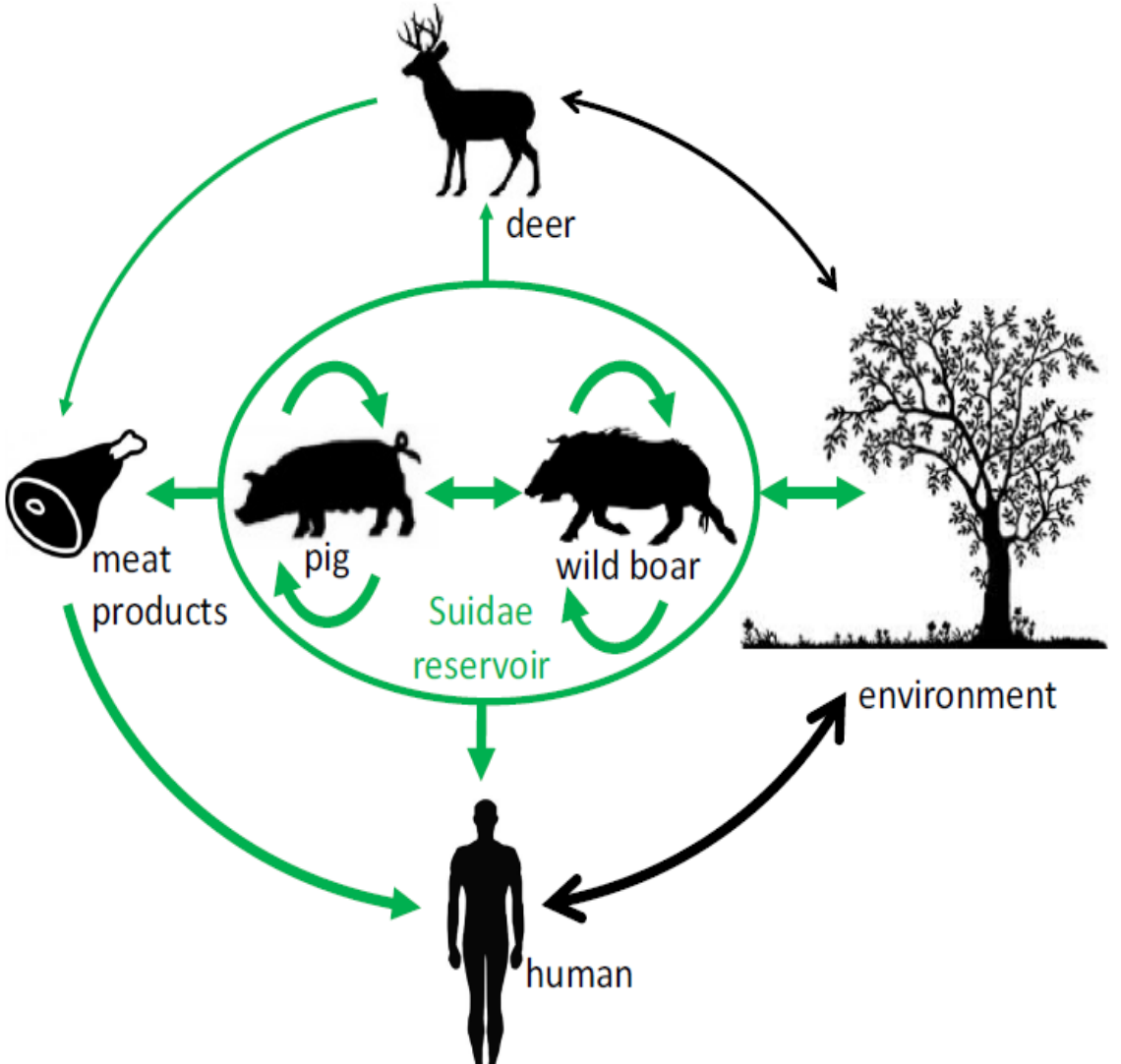
Transmission

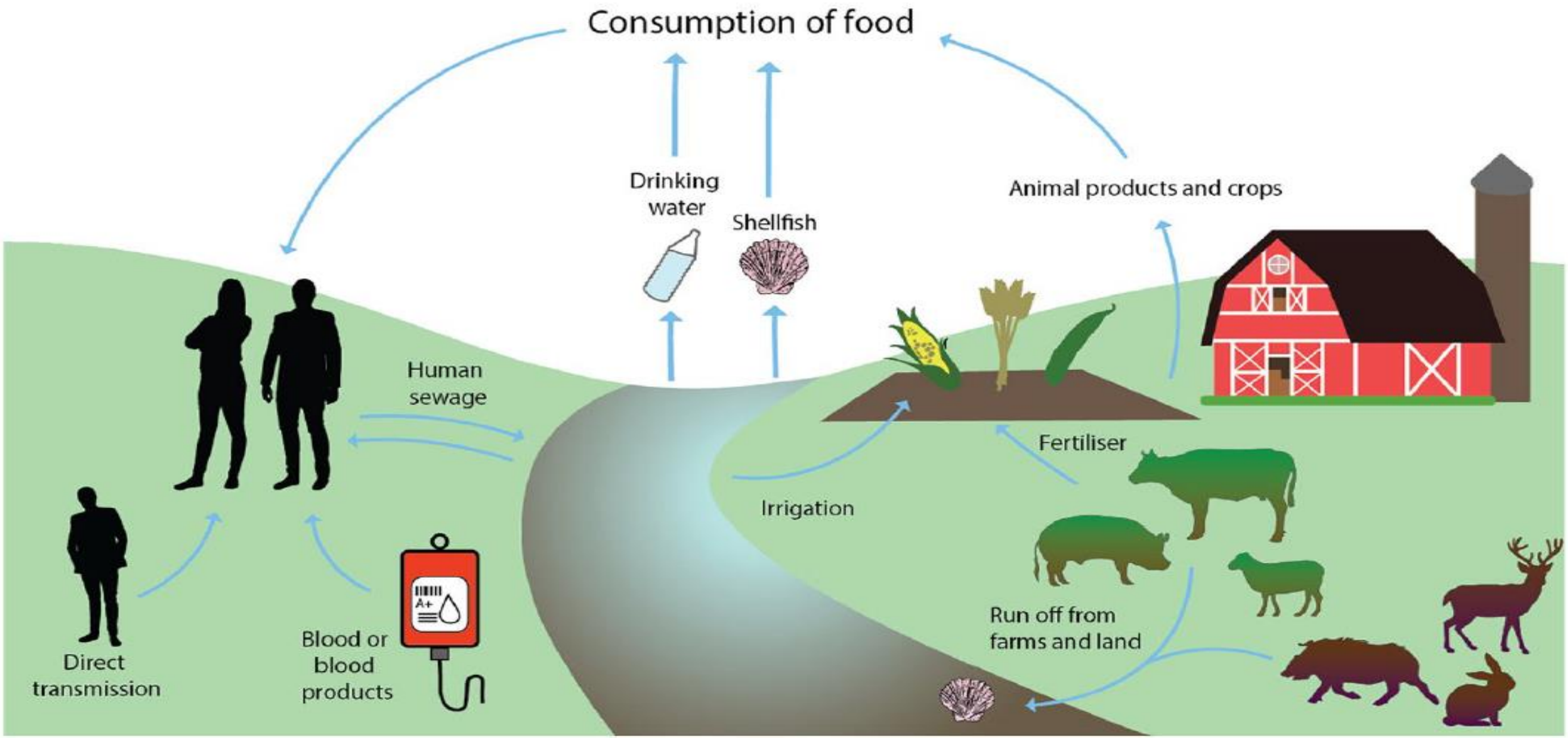
HEV: in tutto il mondo, più comune in Asia orientale e meridionale. HEV trasmesso tramite oro-fecale (acqua contaminata). HEV 1, 2 e 4 relativi alle scadute condizioni igienico-sanitarie in Asia, Africa e America centrale.

- **In Europa, l'HEV è una malattia zoonotica (serbatoio suini HEV3).**
  Prodotti alimentari contaminati (carne di maiale poco cotta, carne di selvaggina, ecc.)

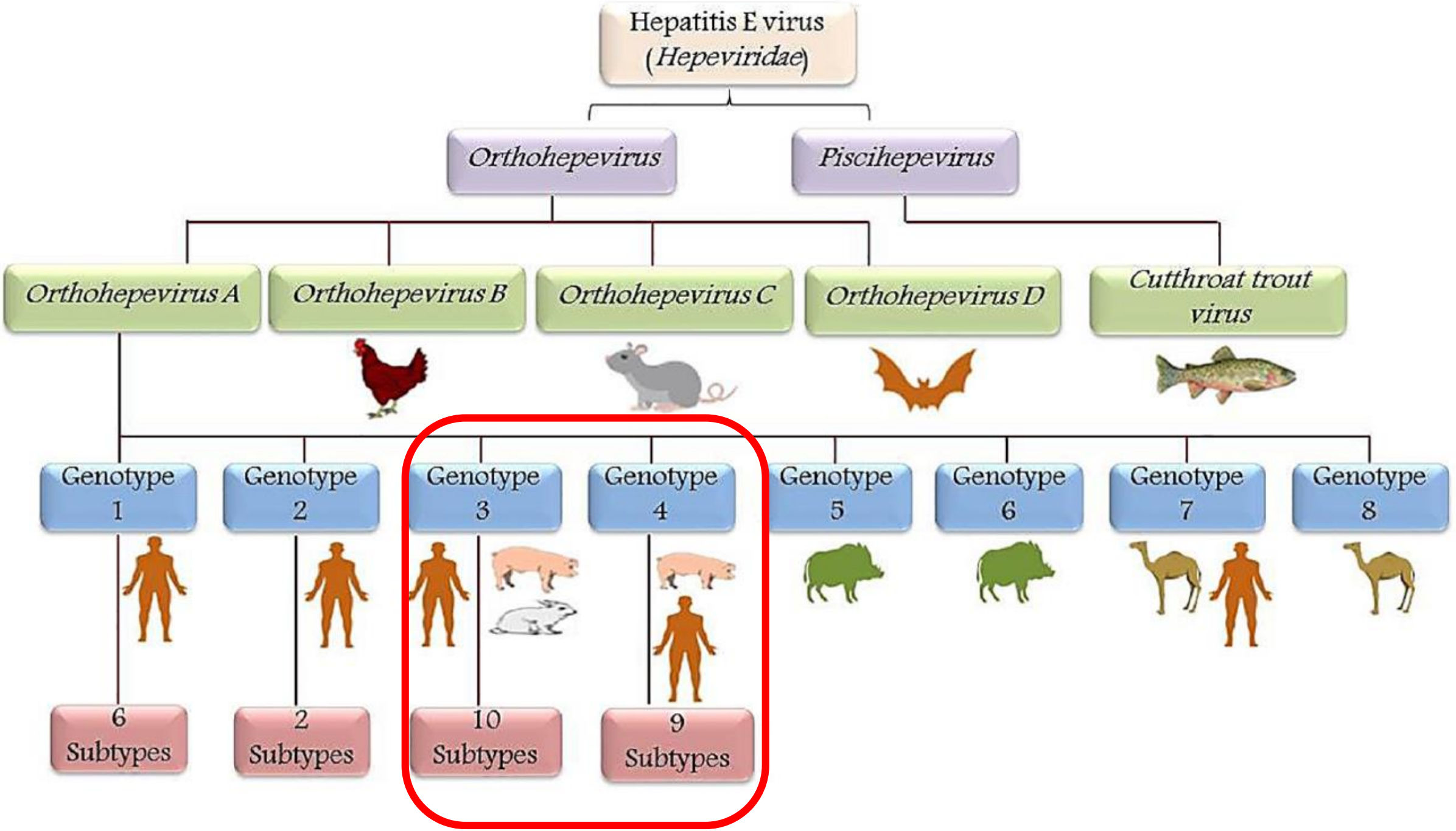
- **Gruppo professionale: lavoratori del macello e silvicoltura, cacciatori, agricoltori, veterinari.**

Le differenze di prevalenza tra/all'interno dei paesi riflettono la diversa esposizione all'HEV, una minore o maggiore endemicità all'interno della popolazione di origine suini, diverse abitudini di consumo (fegato di maiale crudo).





HEV1 and 2 spread by fecal-oral route via contaminated water.
HEV3 and 4 infect humans, pigs, animal species, responsible for sporadic cases


Hepatitis E virus
(Hepeviridae)
Orthohepevirus
Piscihepevirus
Orthohepevirus A
Orthohepevirus B
Orthohepevirus C
Orthohepevirus D
Cutthroat trout virus
Genotype 1
Genotype 2
Genotype 3
Genotype 4
Genotype 5
Genotype 6
Genotype 7
Genotype 8
6 Subtypes
2 Subtypes
10 Subtypes
9 Subtypes

The Global Epidemiology of HEV genotype

Cells **2021**, 10, 2281 3

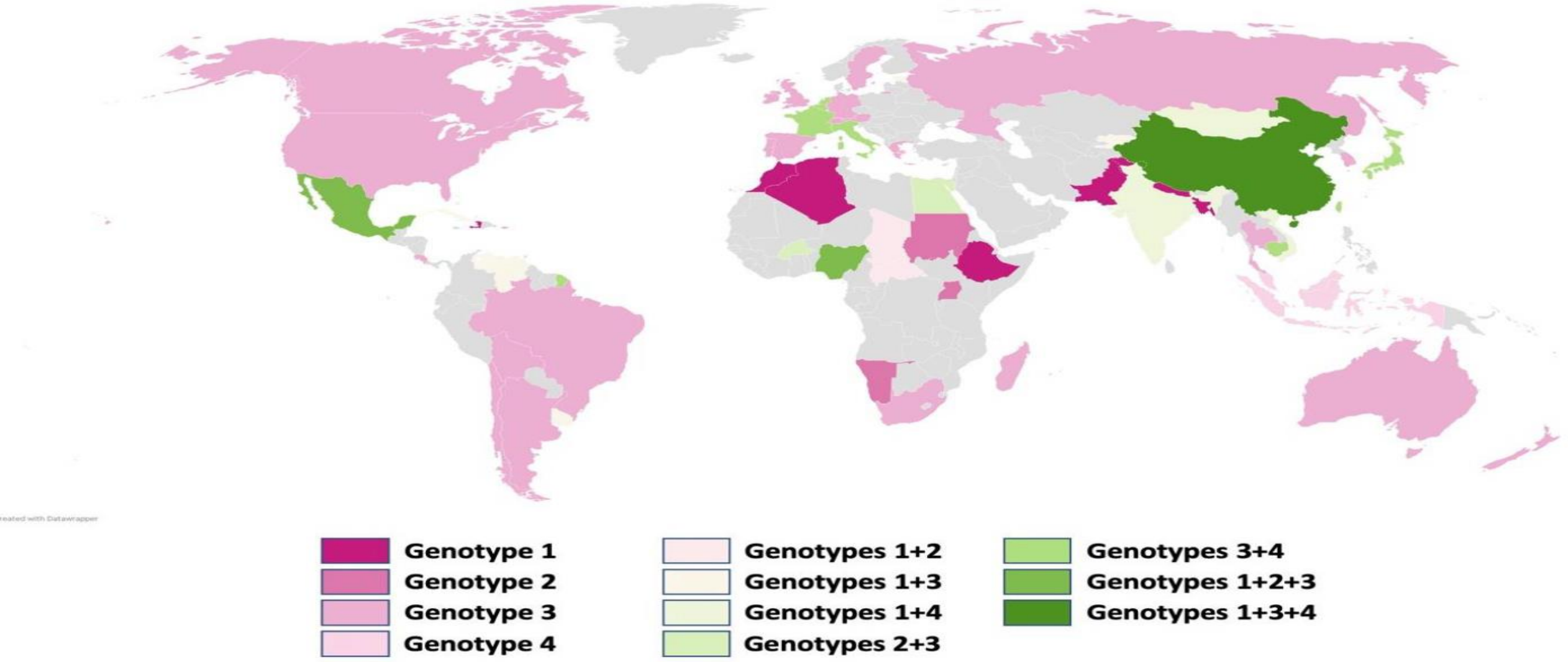


Figure 1. Map with the geographical distribution of the four major human pathogen HEV genotypes. Data adapted from WHO and map generated with Datawrapper, Berlin, Germany.

HEV genotype

Characteristics	Genotype 1	Genotype 2	Genotype 3	Genotype 4
Geographic Location	Africa and Asia	Mexico, West Africa	Developed Countries	China, Taiwan, Japan
Transmission route	Waterborne fecal-oral person-to-person	Waterborne fecal-oral	Food-borne	Food-borne
Groups at high risk for infection	Young Adults	Young Adults	Older Adults (>40 years) and males, immunocompromised persons	Young Adults
Zoonotic transmission	No	No	Yes	Yes
Chronic Infection	No	No	Yes	No
Occurrence of Outbreaks	Common	Smaller scale outbreaks	Uncommon	Uncommon

chttps://www.cdc.gov/hepatitis/hev/hevfaq.htm#section1

Transfusion or transplantation transmitted HEV have sporadically, asymptomatic infection among blood donors is widespread.

- **In last decade, increasing incidence HEV3-donations in European countries.**
- **England and Ireland: selective or universal screening of blood donations.**

TABLE

Prevalence of hepatitis E virus RNA positive donations, population of transplanted patients at risk, reported cases of transfusion-transmitted hepatitis E virus and screening of blood donations in 11 European countries

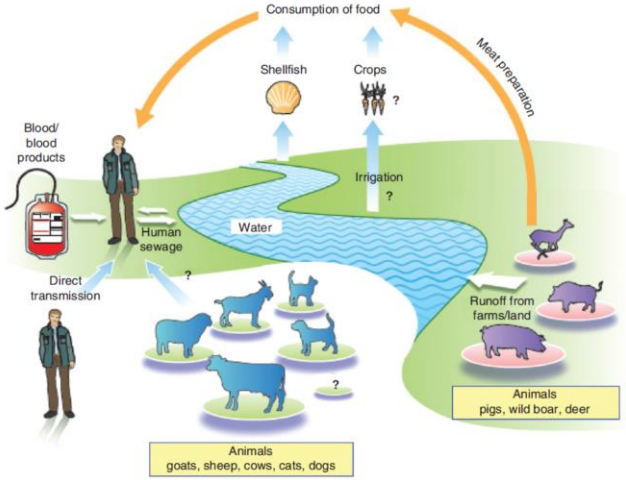
Country	HEV RNA positive donations	Population at risk		Reported TT HEV infections	Screening of blood donations			
		allo-HSCT [51] AN (AR/p1omp)	SOT [52] AN (AR/pmp)		Implemented	Under Consideration	In evaluation	Not recommended
Denmark	1:2,331(2016) [16]	144 (201 – 300)	356 (63.6)					x
France	1:2,218 (2012–3) [18]	1,724 (201 – 300)	5,141(79.6)	x		x[a]		
Germany	1:1,241 (2012) [24]	2,892 (›300)	3,710 (44.9)	x		x[b]		
Greece	NA	169 (151 – 200)	171 (15.4)				x	
Ireland	1:2,778 (2016)	77(151 – 200)	246 (52.3)		x[c]			
Italy	NA	1,625 (201 – 300)	3,252 (53.2)				x	
The Netherlands	1: 726 (2016) [7]	1175 (›300)	1,315 (78.3)			x [d/e]		
Portugal	NA	137 (101 – 150)	739 (69.7)				x	
Spain	1:3,333 (2014) [53]	1,072 (201 – 300)	4,247 (90.2)	x			x	
Switzerland	NA	191 (201 – 300)	504 (61.5)			x		
United Kingdom	1:1,340–5,000 (2016)	1,602 (201 – 300)	4,561 (71.8)	x	x[e/f]			

allo-HSCT: allogeneic haematopoietic stem cell transplant patients; AN: annual number; AR: annual rate; HEV: hepatitis E virus; NA: not

EUROROUNDUP

Hepatitis E and blood donation safety in selected European countries: a shift to screening?

D Domanović [1], R Tedder [2], J Blümel [3], H Zaaijer [4], P Gallian [5], C Niederhauser [6], S Sauleda Oliveras [7], J O'Riordan [8], F Boland [8], L Harritshøj [9], MSJ Nascimento [10], AR Ciccaglione [11], C Politis [12], C Adlhoch [1], B Flan [13], W Oualikene-Gonin [14], G Rautmann [15], P Strengers [16], P Hewitt [17]



HEV prevalence in Italian general population in 6/20 regions:1.5-2.9%. (before 1999)

- **2007-2016: 144 cases of AHE to Italian Surveillance System AVH: 122 (84.7%) M, mean age 40 yrs, 81 (56.6%) Italian.**
- **2004-2016 virological surveillance of 139 AVH non-A, non-B, non-C: 48 (34.5%) HEV. HEV-1 55% (22/40), HEV3 45% (18/40).**
- **2014 HEV-ab blood donor: Abruzzo 48.9% (153/313), Lazio 9.0% (9/100). In Lazio in donor population is higher than general population (2.6% in 1996, 2.9% in 2007)**
- **La prevalenza è aumentata con l'età e associata a salsicce di fegato di maiale essiccate crude. Tra i 153 donatori di sangue IgG positivi in Abruzzo, 2 (1,3%) con IgM e 2 (1,3%) HEV RNA; HEV3c.**

Screening degli emoderivati???

- **Regno Unito su 225000 donazioni di sangue lo 0,035% dei donatori era viremico. Il 42% dei riceventi trasfusi con emoderivati HEVRNA-positivi è diventato viremico o ha sviluppato anticorpi contro l'HEV.**
- **Attualmente, gli emoderivati vengono testati di routine per HEVRNA nel Regno Unito, Irlanda e Paesi Bassi.**
- **Nel Regno Unito si è mostrato l'efficacia in termini di costi dello screening di routine dell'HEV nei trapiantati di organi solidi mediante i test di amplificazione dell'acido nucleico (NAAT) o il test dell'antigene.**
- **Lo screening degli emoderivati plasmatici in US potrebbe non essere necessario poiché lo 0,002% delle donazioni di plasma è HEVRNA positivo.**
- **Lo screening selettivo: Germania e Francia per i pazienti ad alto rischio.**

Le autorità in Grecia, Portogallo, Italia e Spagna stanno valutando se avviare lo screening dell'HEV.

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https://doi.org/10.1007/s12560-021-09461-5

REVIEW PAPER

The Foodborne Transmission of Hepatitis E Virus to Humans

Samantha Treagus[1,3] · Conal Wright[2] · Craig Baker-Austin[3] · Ben Longdon[1] · James Lowther[3]

Table 1 Pattern of infection for the different genotypes of HEV, adapted from Centers for Disease Control and Prevention (2020)

Genotype	Transmission in humans?	Transmission routes	Geographical distribution pattern	Extrahepatic manifestations	Age groups at higher risk	Gender more commonly affected	Lethality
1	Yes	Faecal-oral; waterborne; blood transfusion; organ donation	Economically developed and developing countries	Pancreatic	Differs by country[a,b]	Differs by country[a,b]	0.5–1%[c]; 20% in pregnant women[d,e,f]
2	Yes	Faecal-oral; waterborne; blood transfusion; organ donation	Economically developing countries	Unknown	Young adults	Unknown	0.5–1%[c]
3	Yes	Foodborne; blood transfusion; organ donation	Economically developed and developing countries	Chronic infections in immune-compromised patients. Neurological, haematological, immunological and renal manifestations[g]	Older adults (>40 years)	Males	0.5–1%[c]
4	Yes	Foodborne; blood transfusion; organ donation	Economically developed and developing countries	Unknown	Young adults	Possibly males (limited data)[h]	0.5–1%[c]
5	No	Faecal-oral	Unknown	Unknown	Unknown	Unknown	Unknown
6	No	Faecal-oral	Unknown	Unknown	Unknown	Unknown	Unknown
7	Yes	Foodborne; Faecal-oral; blood transfusion?	Unknown	Unknown	Unknown	Unknown	Unknown
8	No	Faecal-oral	Unknown	Unknown	Unknown	Unknown	Unknown

Table 3 A list of the seroprevalence levels of anti-HEV antibodies in pigs and the percentage of pork products found to be HEV positive by RT-PCR in those locations

Location	Pig sample population anti-HEV antibody seroprevalence	Percentage of tested foodstuffs HEV positive by RT-PCR
Brazil	63.6% of 357 pigs (Vitral et al. 2005)	1.7% of 118 slaughterhouse livers (Gardinali et al. 2012)
Canada	59.4% of 998 pigs (Yoo et al. 2001)	8.8% of 283 livers, 1.0% of 599 pork chops (Wilhelm et al. 2014)
		47.0% of 76 pork pâtés and 10.5% of 19 retail raw pork livers (Mykytczuk et al. 2017)
France	31.0% of 6565 pigs (Rose et al. 2011); 60% of 1034 pigs (Feurer et al. 2018)	4.0% of 3715 slaughterhouse livers (Rose et al. 2011)
		2.8% of 1034 slaughterhouse livers (Feurer et al. 2018)
		30.0% of 140 figatelli and fitone, 29.0% of 169 liver sausages, 25.0% of 55 quenelles or quenelle paste, 3.0% of 30 dried salted livers (Pavio et al. 2014)
		58.3% of 12 raw liver sausage (Colson et al. 2010)
Germany	49.8% of 1072 pigs (Baechlein et al. 2010)	4.0% of 200 retail livers (Wenzel et al. 2011)
		20.0% of 70 raw sausages and 22.0% of 50 liver sausages (Szabo et al. 2015)
Italy	45.1% of 2700 pigs (Mughini-Gras et al. 2017)	20.8% of 48 slaughterhouse livers (Di Bartolo et al. 2011)
		6.0% of 33 slaughterhouse livers (Di Bartolo et al. 2012)
		13.3% of 15 fresh liver sausages, 7.1% of 14 dried liver sausages (Di Bartolo et al. 2015)
Japan	57.9% of 2500 pigs (Takahashi et al. 2003)	1.9% of 363 retail livers (Yazaki et al. 2003)
Netherlands	89.0% of 417 organic pigs, 72% of 265 conventionally farmed pigs, 76% of 164 free range pigs (Rutjes et al. 2014)	6.5% of 62 commercial pork livers (Bouwknegt et al. 2007)
		12.7% of 79 livers, 70.7% of 99 liver sausages, 68.9% of 90 liver pâté samples (Boxman et al. 2019)
Spain	20.4% of 1441 pigs (de Oya et al. 2011)	6.0% of 93 sausages, 3.0% of 39 slaughterhouse livers (Di Bartolo et al. 2012)
Switzerland	62.3% of 1001 pigs in 2006, 53.8% of 999 pigs in 2011 (Burri et al. 2014)	1.3% of 160 slaughterhouse livers (Müller et al. 2017)
		11.8% of 102 raw liver sausages (Giannini et al. 2017)
		11.7% of 90 pork liver and raw meat sausages (Moor et al. 2018)
UK	92.8% of 629 pigs, with 20.5% viraemic at slaughter (Grierson et al. 2015)	10.0% of 63 sausages, 3.0% of 40 slaughterhouse livers (Berto et al. 2012)
USA	21.9% of 182 pigs (Owolodun et al. 2013)	11.0% of 127 retail liver (Feagins et al. 2007)

Table 8 The presence of HEV in shellfish in different countries

Location	Study	Percentage of shellfish HEV positive
China	Gao et al. (2015)	17.5% of 126 shellfish samples[a] of various species from production areas
Denmark	Krog et al. (2014)	0% of 29 mussel samples[a] from 19 production areas
France	Grodzki et al. (2014)	0% of 286 shellfish samples[a] of various species from two production areas
Italy	La Rosa et al. (2018)	2.6% of 384 shellfish samples[a] of various species from production areas
Japan	Li et al. (2007)	6.3% of 32 Yamato-Shijimi clam samples[a]
Scotland	Crossan et al. (2012)	85.4% of 48 individual wild mussels
	O'Hara et al. (2018)	2.9% of 310 retail shellfish samples[a] (mussels and oysters)
Spain	Mesquita et al. (2016)	14.8% of 81 mussel samples[a] from a production area
	Rivadulla et al. (2019)	24.4% of 164 mussel, clam, and cockle samples[a]

[a]Where the study states that samples of shellfish were tested, it was either stated or assumed in each publication that each "sample" would have been formed by ten or more shellfish individuals and is therefore technically a pooled sample

Case-based hepatitis E surveillance data by number of collecting EU/EEA countries, 2005–2015

Case information collected	Number of countries
Date of notification to surveillance organisation	20
Sex	20
Age or date of birth	19
Date of clinical onset	19
Source of notification	19
Travel history within EU/EEA	17
Unique patient identifier	17
Travel history outside EU/EEA	16
Date of diagnosis	15
Hospitalisation	15
Occupation	15
HEV-related death	14
Clinical symptoms	13
Cluster link	10
Environmental contact with livestock/farm animals	9
Pregnancy	9
Food consumption history – groups of foods	8
Food consumption history – detailed food items	7
Ethnicity	6
Immunosuppressive medication or condition	6
Recent transfusion of blood components or blood products	6
Alcohol consumption	5
Medication	5
Other underlying medical conditions	5
Other	5
Recent transplantation	5
Migration background/refugee status	2

Data from the ECDC surveillance study in EU/EEA countries [6,22].

foods

MDPI

Article

Quantitative Methods for the Prioritization of Foods Implicated in the Transmission of Hepatititis E to Humans in Italy

Ornella Moro [1,2,*], Elisabetta Suffredini [1], Marco Isopi [2], Maria Elena Tosti [3], Pietro Schembri [4] and Gaia Scavia [1]

…we developed a mathematical model to rank importance of various types of food implicated in transmission of HEV to humans in adult Italian population

(around 50,000,000).

The food categories considered are pork products with liver, pork products without liver, bivalve shellfish, green leafy vegetables and raw milk.

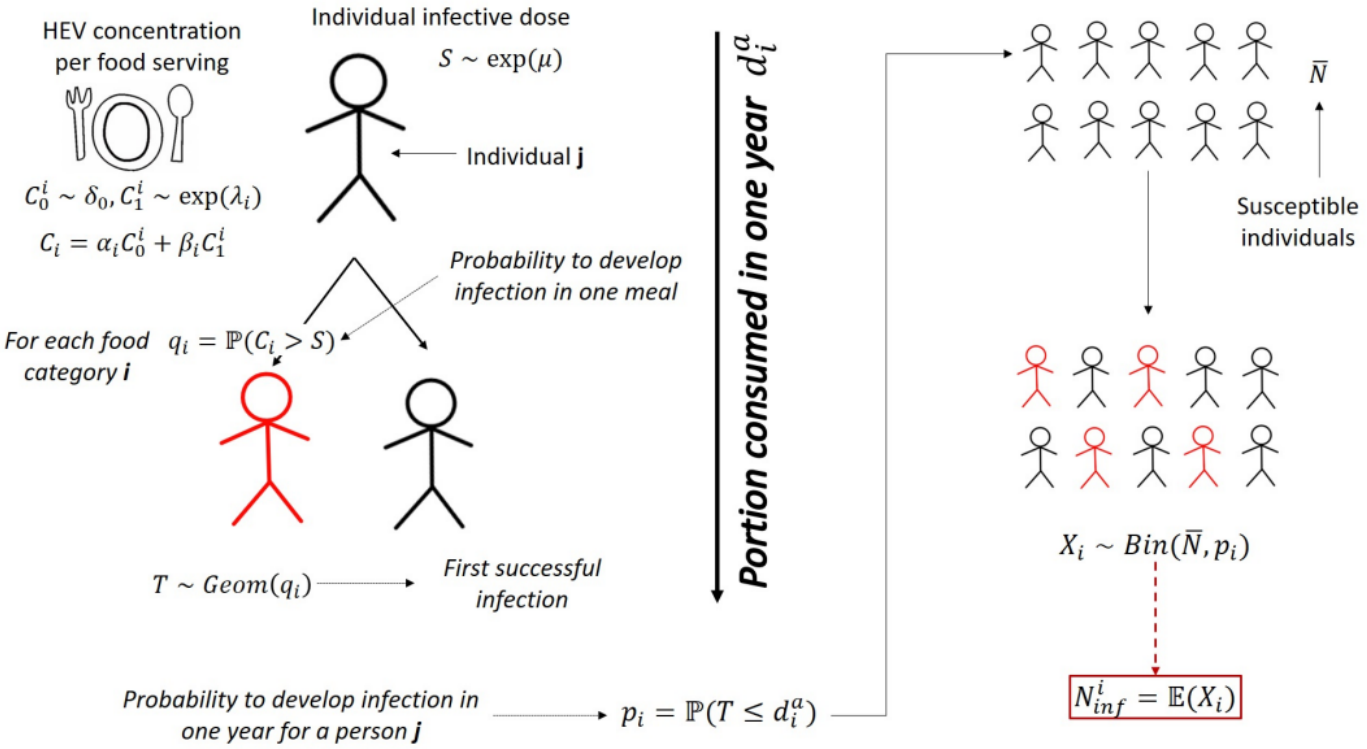

Figure 1. The model sketch.

Table 1. Model parameters.

foods

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to the Italian surveillance system for acute viral hepatitis (SEIEVA).

The food sampling survey indicated the highest proportion of HEV positive samples (i.e., HEV food prevalence αi) belongs to pork products with liver (11%), followed by pork products without liver (2.8%) and bivalve shellfish.

No positive samples were found for green leafy vegetables and raw milk.

shellfish	186	49	26
wild boar meat	77	17	22
vegetables	34	5	15
wild boar cured meat	55	8	14
offal	53	6	11
other game meat	53	4	7

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Article

Occurrence of Human Enteric Viruses in Shellfish along the Production and Distribution Chain in Sicily, Italy

Giusi Macaluso [1,†], Annalisa Guercio [1,†], Francesca Gucciardi [1,*], Santina Di Bella [1], Giuseppina La Rosa [2], Elisabetta Suffredini [3], Walter Randazzo [4] and Giuseppa Purpari [1]

- **Aim: prevalence of enteric virus contamination along shellfish production and distribution chain in Sicily.**
- 162 batches of mollusks between 2017 and 2019 from harvesting areas, depuration and dispatch centers (n = 63), restaurants (n = 6) and retail stores (n = 93) distributed all over the island.
- **Results: 5.56% of samples were contaminated with at least one NoV, HAV and/or HEV.** I molluschi contaminati sono stati campionati nei siti di produzione e nei negozi al dettaglio e la loro origine risale alla Spagna e a diversi comuni italiani.
- Our study highlights **the need to implement routine monitoring programs along the whole food chain** as an effective measure **to prevent foodborne transmission of enteric viruses.**

Sampling sites selected to monitor the levels of enteric virus contamination in shellfish along the Sicilian food chain, 2017-2019.

foods

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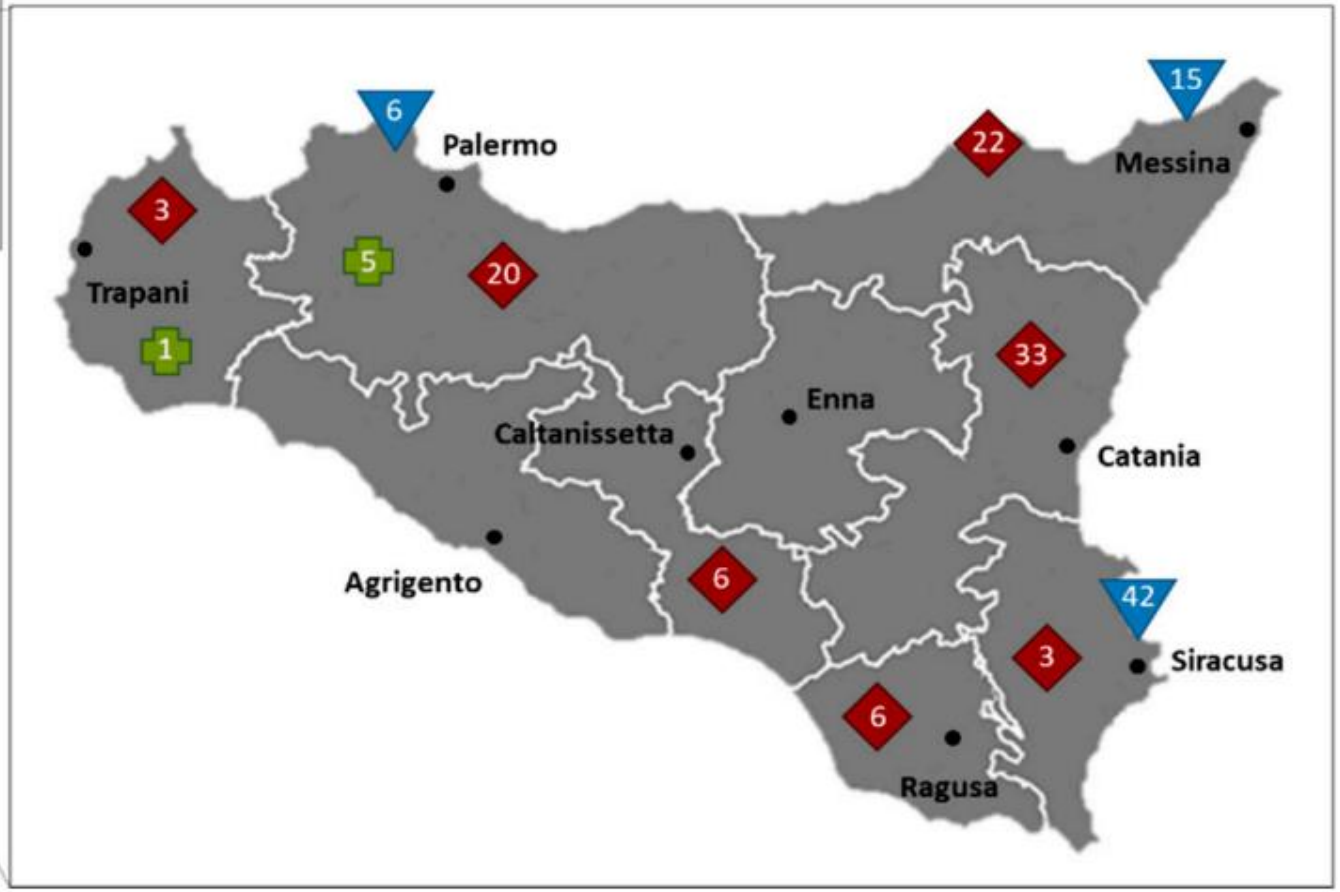


Harvesting areas, depuration, and shipping centers

Restaurants

Shellfish markets and retail stores

RISCHIO…..

l'infezione da HEV nel suino evolve in maniera asintomatica (Clemente- *Casares et al., 2003; Banks et al., 2004a; Zheng et al., 2006; Fernandez-Barredo et a., 2006).*

HEV in animali in fase di macellazione, o nella fase immediatamente precedente alla commercializzazione dei prodotti carnei di origine animale: 64,6% (31\48 *suini) sono positivi HEVRNA*.

Nei suini giovani (3-4 mesi età): la prevalenza era del 95% (19\20), mentre negli adulti (9-10 mesi età): 42,9% (12/28). *(Caprioli et al., 2007; Di Bartolo et al., 2008)*

- **In molti paesi Europei: *animali positivi per HEV sono riscontrati in tutte le fasce d'età*** *(Di Bartolo et al., 2008Fernandez-Barredo et al. 2006; de Deus et al., 2007; Leblanc et al., 2007; McCreary et al., 2008; Breum et al., 2010).*

- **HEVRNA è presente per periodi più lunghi e con *frequenza più elevata nella bile rispetto a fegato, feci e siero*** *(Halbur et al., 2001)* **. In sede di macellazione, la bile rappresenta un importante veicolo di trasmissione per il personale che lavora le carni di maiale, e un'importante fonte di cross- contaminazione.**

Thermal inactivation treatments for HEV in non-food matrix samples

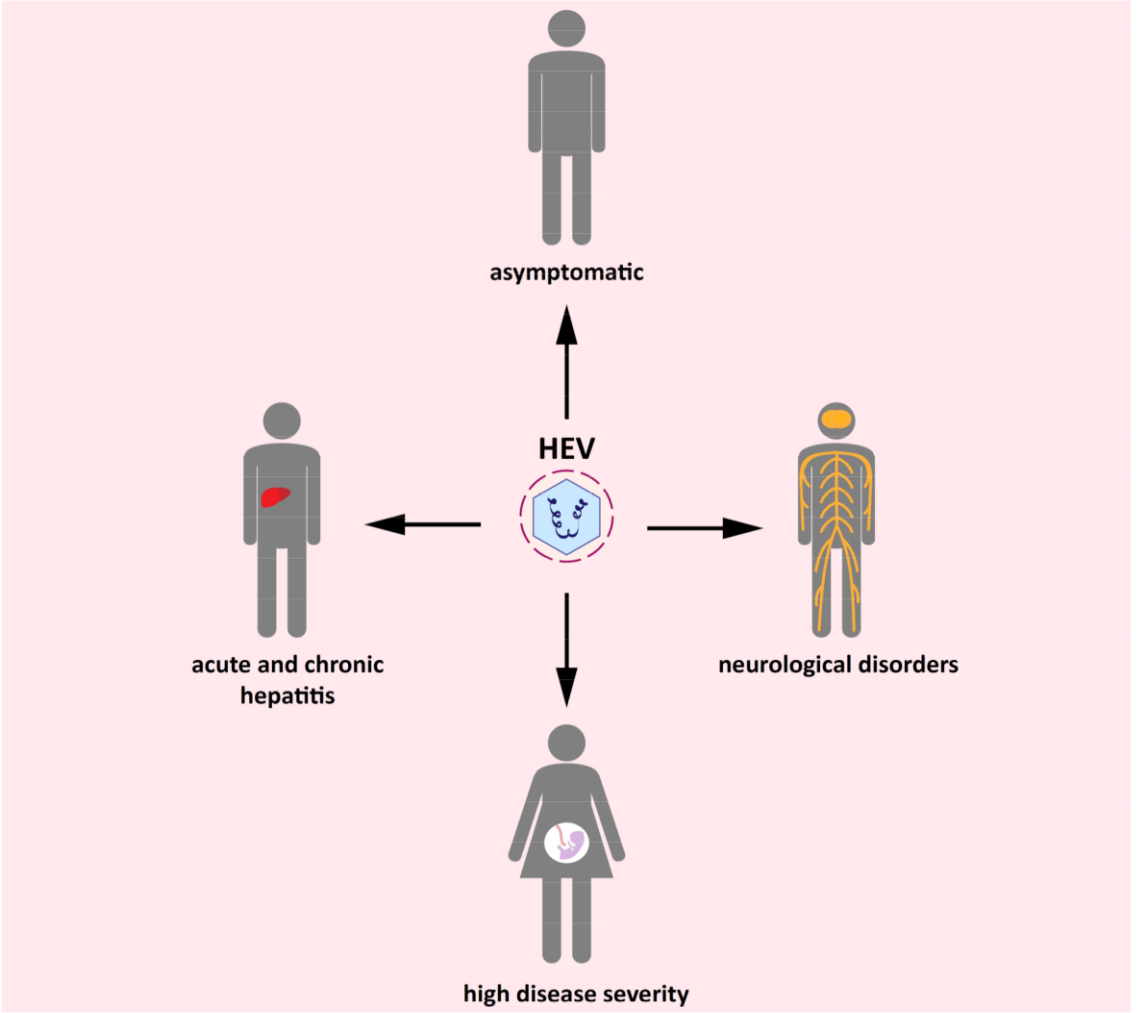
Study	Cell culture or molecular detection?	Genotype	Heat treatment		Growth period	Inactivation/reduction
			Temperature (°C)	Time (min)		
Emerson et al. (2005)	Cell culture, HepG2/C3A cells	1 (strain Akluj[a])	56	60	5–6 days	> 80% reduction
		1 (strain SAR55[b])	56	60	5–6 days	~ 50% reduction
		2 (strain Mex14[c])	60	60	5–6 days	96% reduction
Huang et al. (1999)	Cell culture, A549 cells	3 (strains G93-1*, G93-2*, G93-3*, G93-4*)	56	30	72 h	< 1.0 (TCID50/0.025 ml)
Johne et al. (2016)	Cell culture, A549/D3 cells	3 (strain 47832c[f])	55	1	35 days	~ 1 log reduction in focus forming units
			70	2		"No infectious virus"
Schielke et al. (2011)	Molecular detection	3 (strain wbGER27[e])	56	15	N/A	74.07% reduction
		3 (strain wbGER27[e])	56	60	N/A	99.90% reduction
Tanaka et al. (2007)	Cell culture, PLC/PRF/5 cells	3 (strain JE03-1760F[d])	70	10	35 days	"No infectious virus"
		3 (strain JE03-1760F[d])	56	30	50 days	"Still infectious"

thermal inactivation treatments for HEV in

Study	Food stuff	Cooking method	Temperature (°C)	Time (min)	Measurement of inactivation	HEV inactivated?
Barnaud et al. (2012)	Pâté preparation (spiked with 10^8 HEV genome copies)	Water bath	62	5	Intravenous administration to pigs	No
				20		No
				120		No
			68	5		No
				10		No
						No
						No
						No
				20		Yes
Feagins et al. (2008)	Pig liver (naturally infected)	Incubation	56	60	Oral administration to pigs	No
		Boiling	≥ 71 (internal)	5		Yes
		Stir fry	≥ 71 (internal)	5		Yes
Imagawa et al. (2018)	Minced meat (spiked with 10^{10} HEV genome copies)	Boiling or roasting	63	1	Cell culture	No
				5		No
				30		Yes
			65	1		No
				5		Yes
			70	1		No
				5		Yes

l'HEV può essere inattivato se riscaldato fino a 71 °C per almeno 20 min

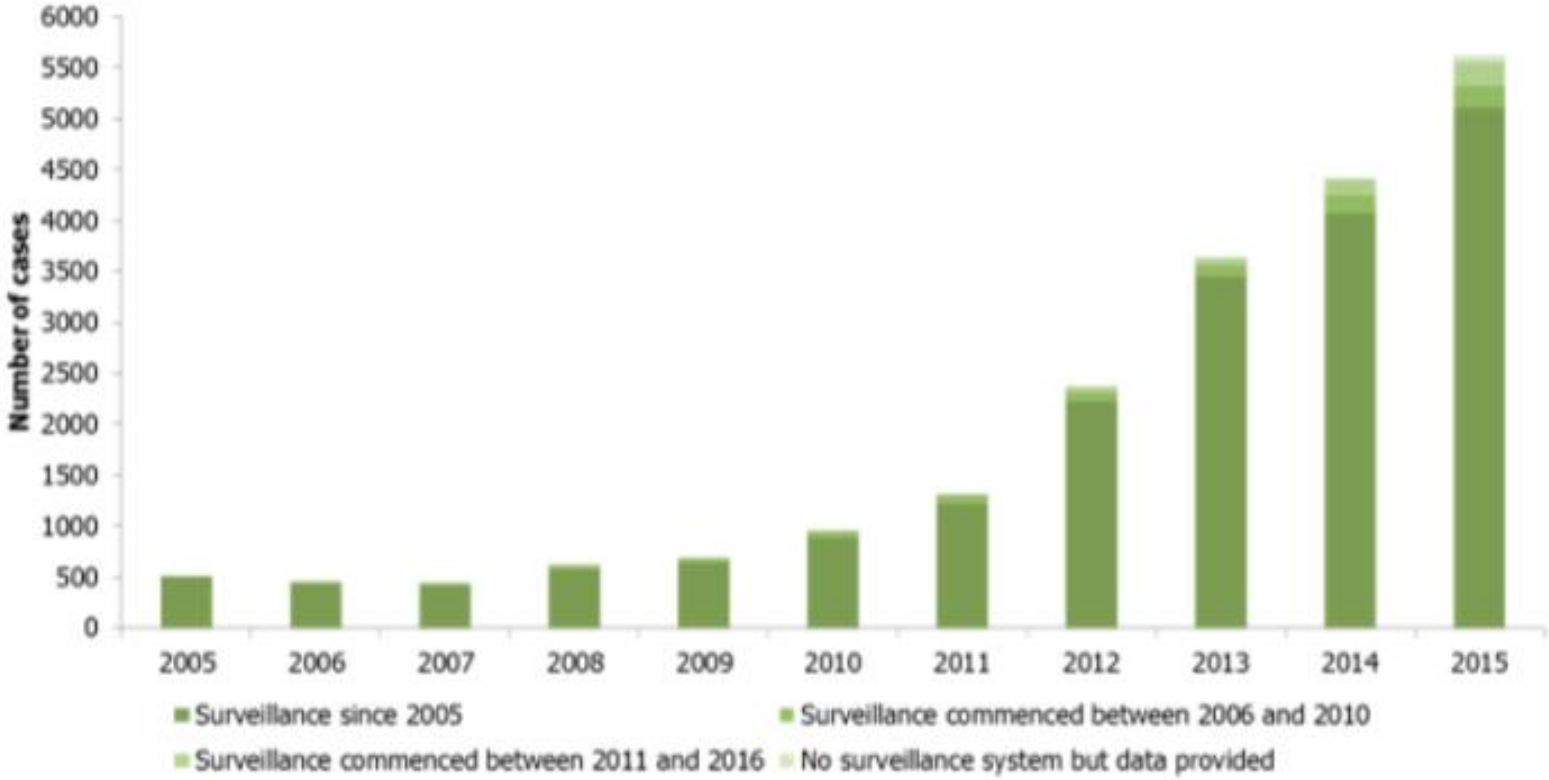
When testing



HEV testing and sequencing of isolates performed in EU/EEA countries as of 2015

ecdc
EUROPEAN CENTRE FOR DISEASE PREVENTION AND CONTROL

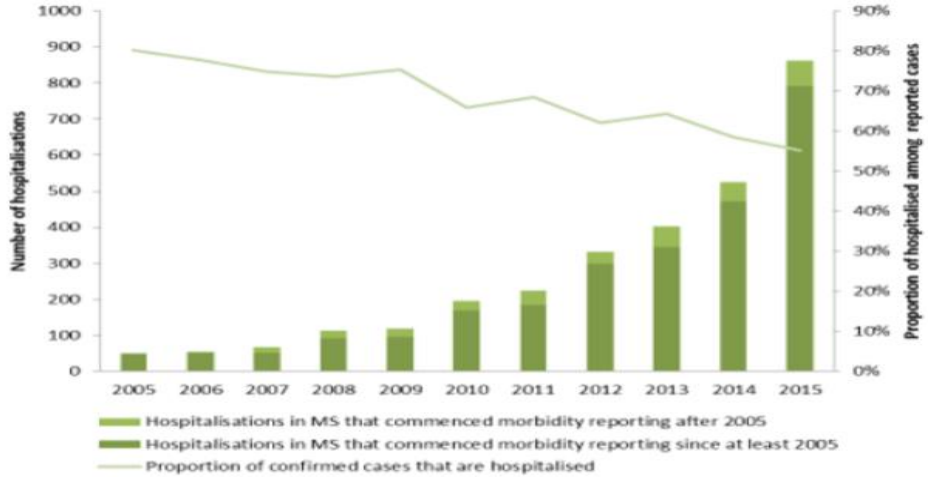
Number of laboratory-confirmed cases of HEV by year and start of surveillance, 22 EU/EEA Member States, 2005–2015*



- 10-fold increase 2005–2015 due locally acquired infections
- 78% of cases reported from France, Germany and UK

* Data available for: Austria, Belgium, Bulgaria, Croatia, Cyprus, Czech Republic, Estonia, Finland, France, Germany, Hungary, Italy, Latvia, Netherlands, Norway, Poland, Portugal, Slovakia, Slovenia, Spain, Sweden, United Kingdom

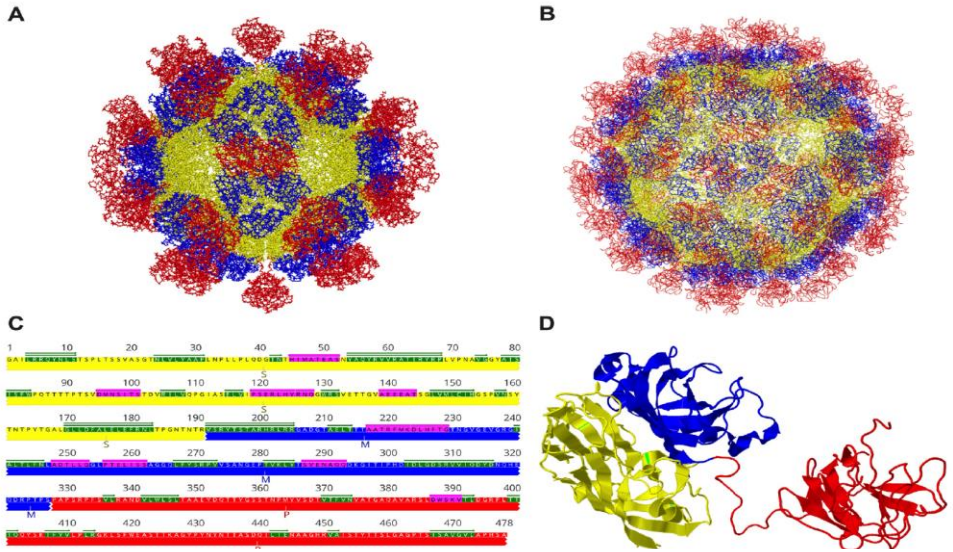
Number of hospitalisations in confirmed HEV cases in 14 EU/EEA Member States, 2005–2015*

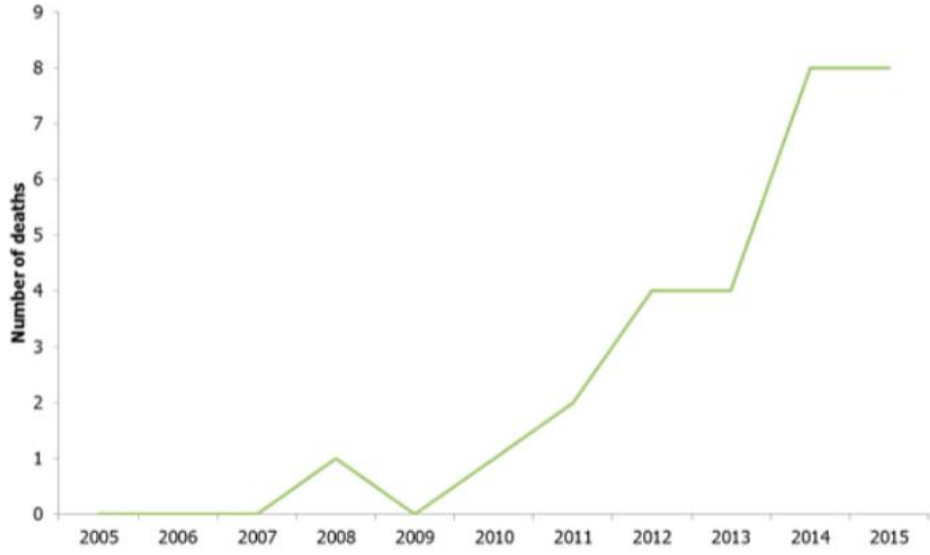


The proportion of hospitalised cases decreased
-> higher awareness in non-hospital settings and inclusion of HEV in routine diagnosis?

* Data on hospitalisation available for: Austria, Belgium, Croatia, Czech Republic, Estonia, Germany, Hungary, Italy, Latvia, Poland, Portugal, United Kingdom (Northern Ireland)

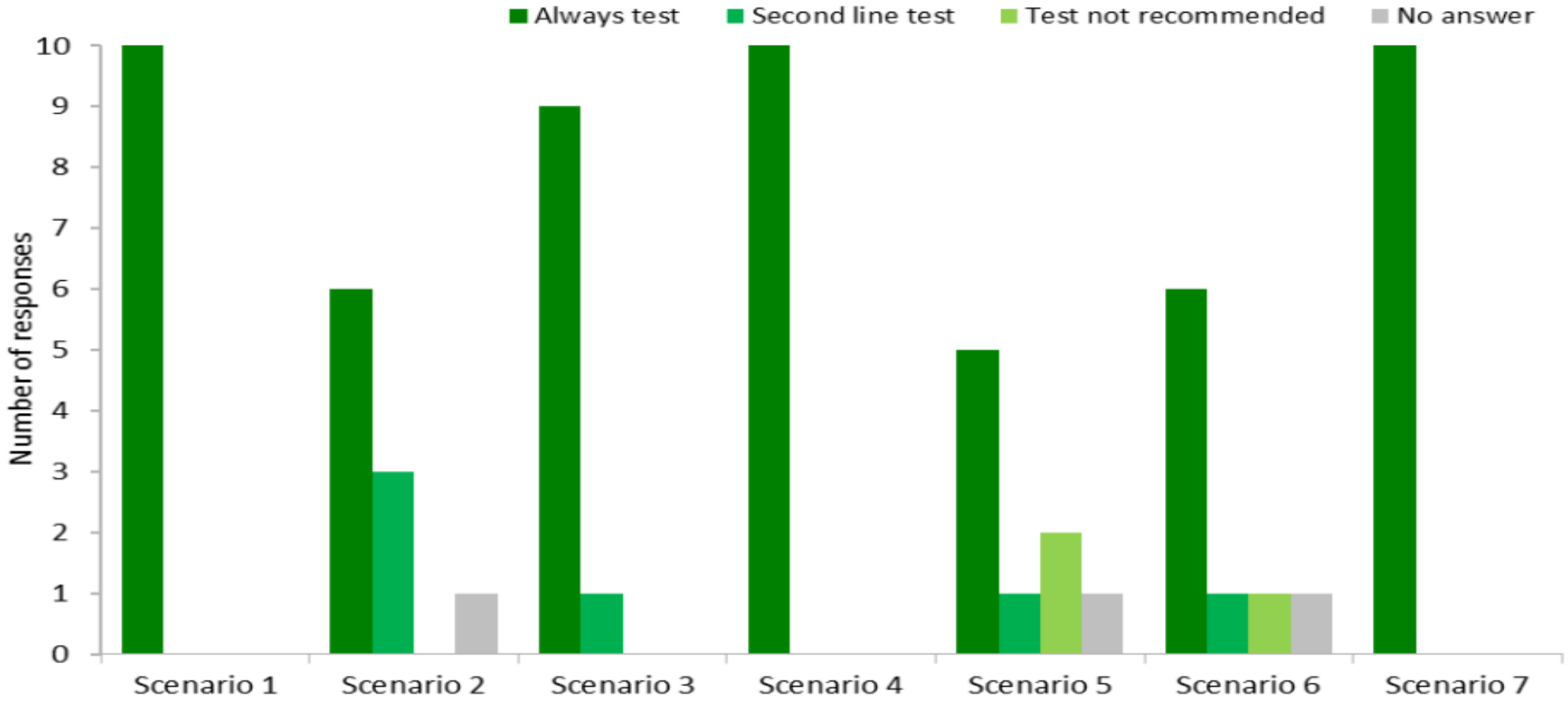
Number of deaths associated with HEV infection, 12 EU/EEA Member States, 2005–2015*





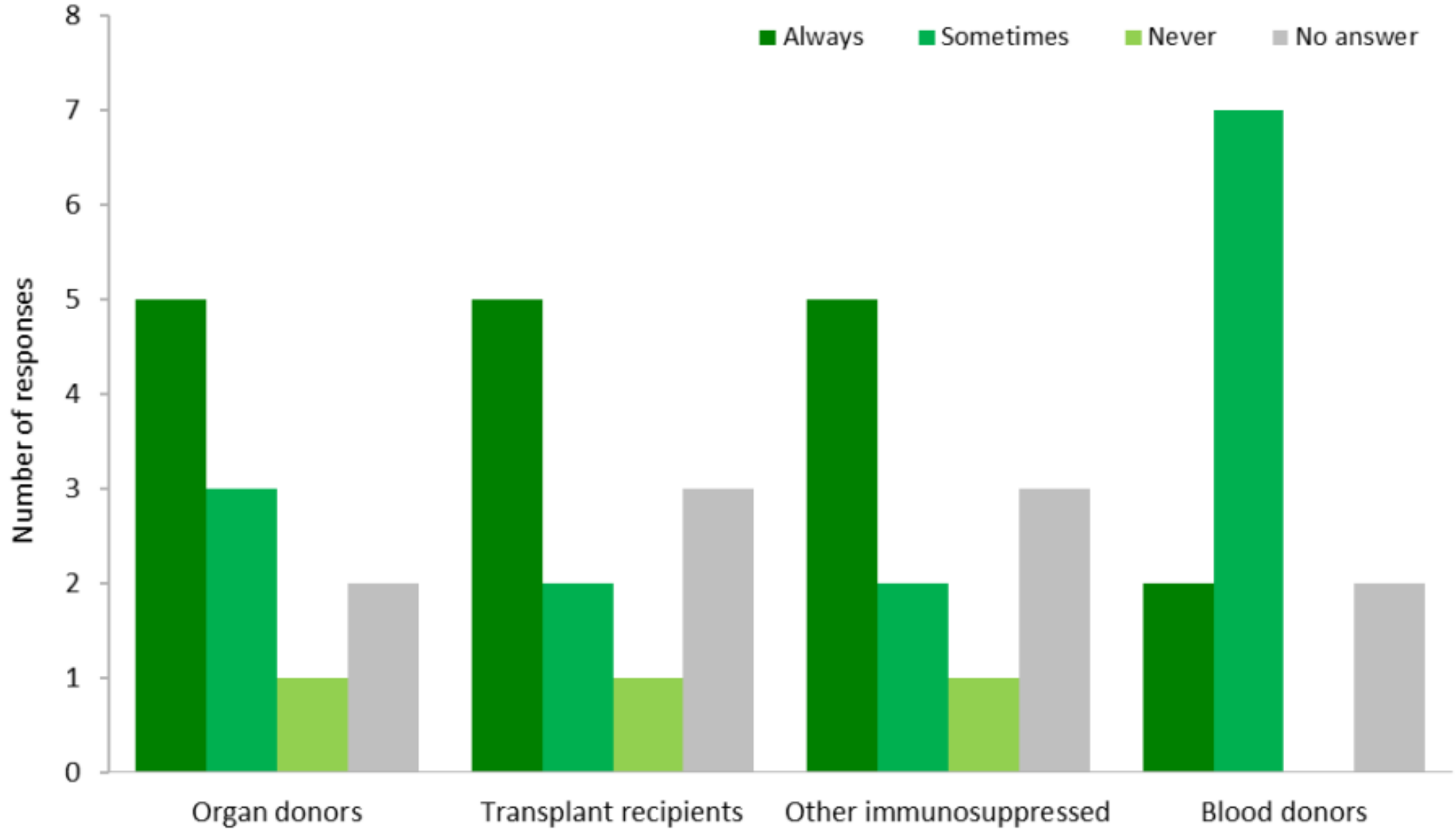
28 deaths associated with HEV infection reported from five countries

Need for HEV testing in high-risk patients



Scenario 1: presentation with symptoms of viral hepatitis, no other information known
Scenario 2: presentation with deranged liver function tests (LFTs) but no symptoms of viral hepatitis
Scenario 3: presentation with deranged LFTs and non-specific symptoms
Scenario 4: presentation with deranged LFTs and symptoms of viral hepatitis
Scenario 5: presentation with epidemiological link but no symptoms
Scenario 6: presentation with epidemiological link and non-specific symptoms
Scenario 7: presentation with epidemiological link and symptoms of viral hepatitis.

Figure 7. Routine testing for HEV in risk groups



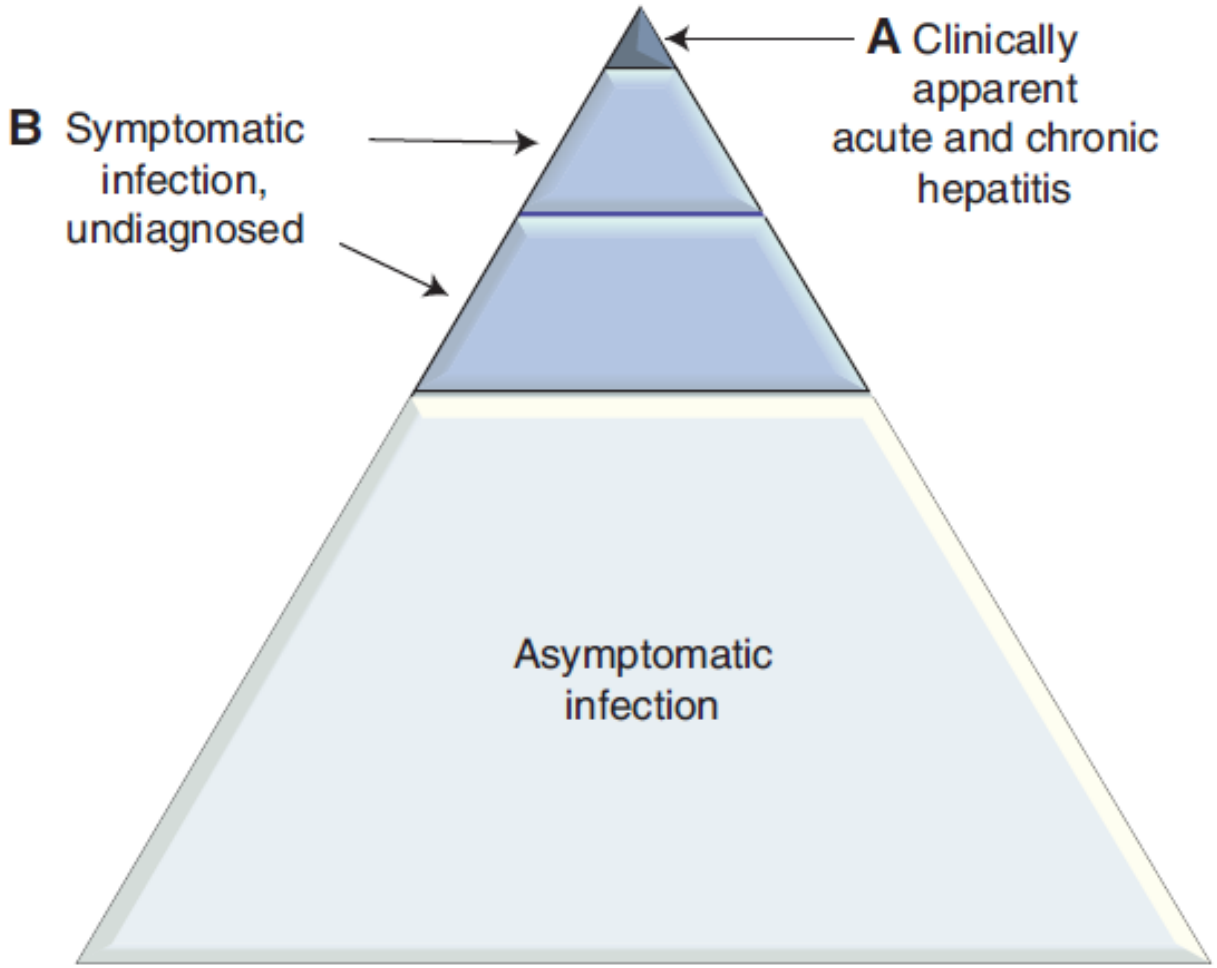
Recommendations and suggestions for HEV testing in clinical patients according to EASL

Manifestation	HEV testing	Level of evidence
Acute	Recommended: All patients with symptoms consistent with acute hepatitis irrespective of travel history	A1
Acute	Recommended: Travellers with hepatitis returning from areas endemic for HEV gt1 or 2	A1
Acute	Recommended: Patients presenting with drug-induced liver injury	A1
Acute	Recommended: Patients with unexplained flares of chronic liver disease	C2
Chronic	Recommended: All immunosuppressed patients with unexplained abnormal LFTs	A1
Extrahepatic	Recommended: Patients presenting with neuralgic amyotrophy irrespective of LFT results	B1
Extrahepatic	Recommended: Patients presenting with Guillain-Barré syndrome irrespective of LFT results	B1
Extrahepatic	Suggested: Patients with encephalitis/myelitis	C2
SoHO	Recommended: Patients with abnormal LFTs after receiving blood products	A1
SoHO	Recommended: blood donor services screen blood donors for HEV by NAT, informed by local risk assessment and cost-effectiveness studies	A1

LFT: liver function test[i]
NAT: nucleic acid amplification test
SoHO: substances of human origin
Level of evidence A: data derived from meta-analyses or systematic reviews or from (multiple) randomised controlled trials (RCTs) with high quality

HEV Presentazione Clinica

- **Il periodo di incubazione: 2-10 wks, (media 5-6 wks).**
- **Le persone infette espellono HEV da pochi giorni prima fino a 3-4wks dopo l'insorgenza della malattia.**
- **Il rapporto tra infezione sintomatica e asintomatica varia da 1:2 a 1:13.**
- **Più del 90% di HEV3 e 4 sono asintomatici. Una piccola minoranza di pazienti (maschi più anziani) sviluppa epatite sintomatica.**



H.R. Dalton and J. Izopet

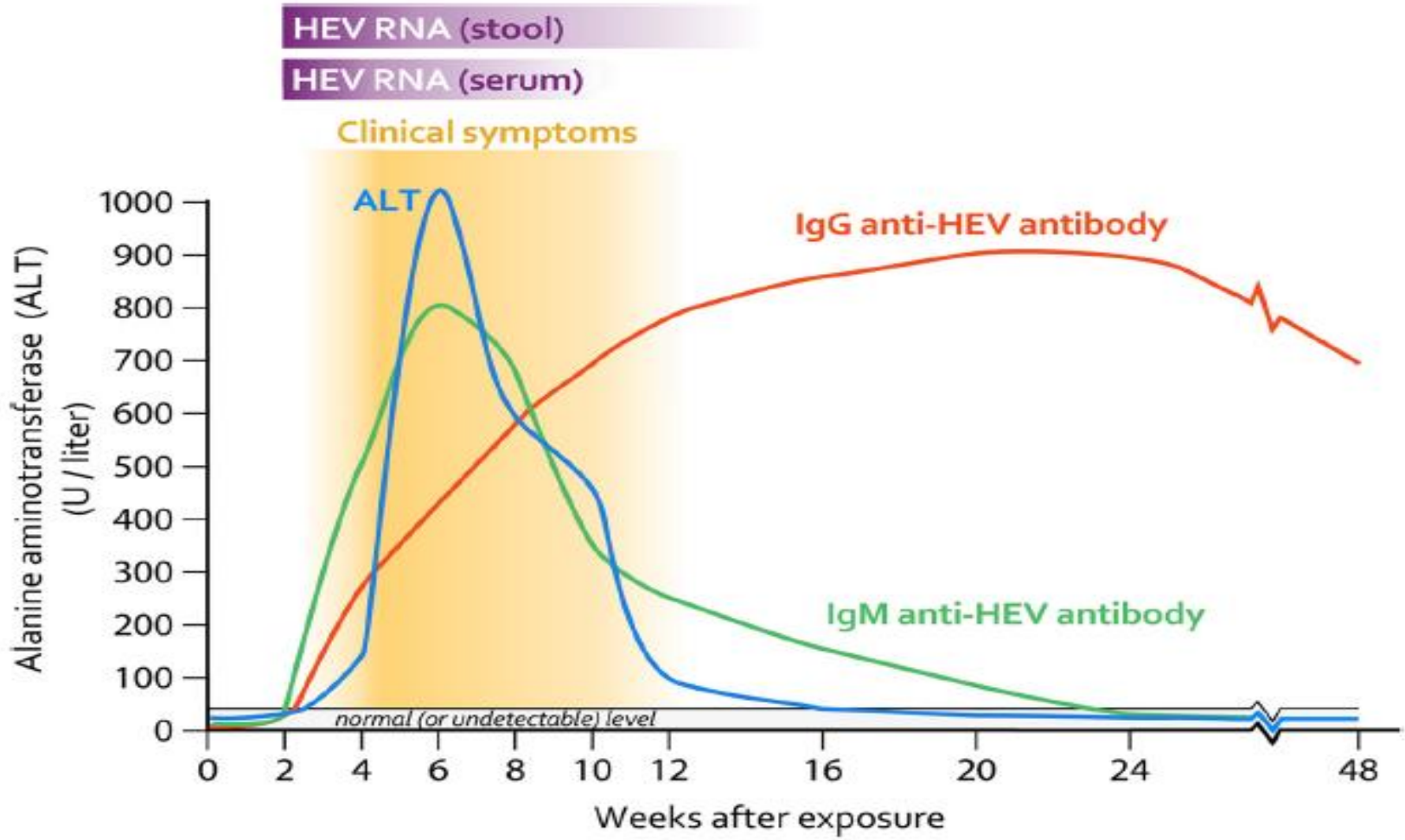
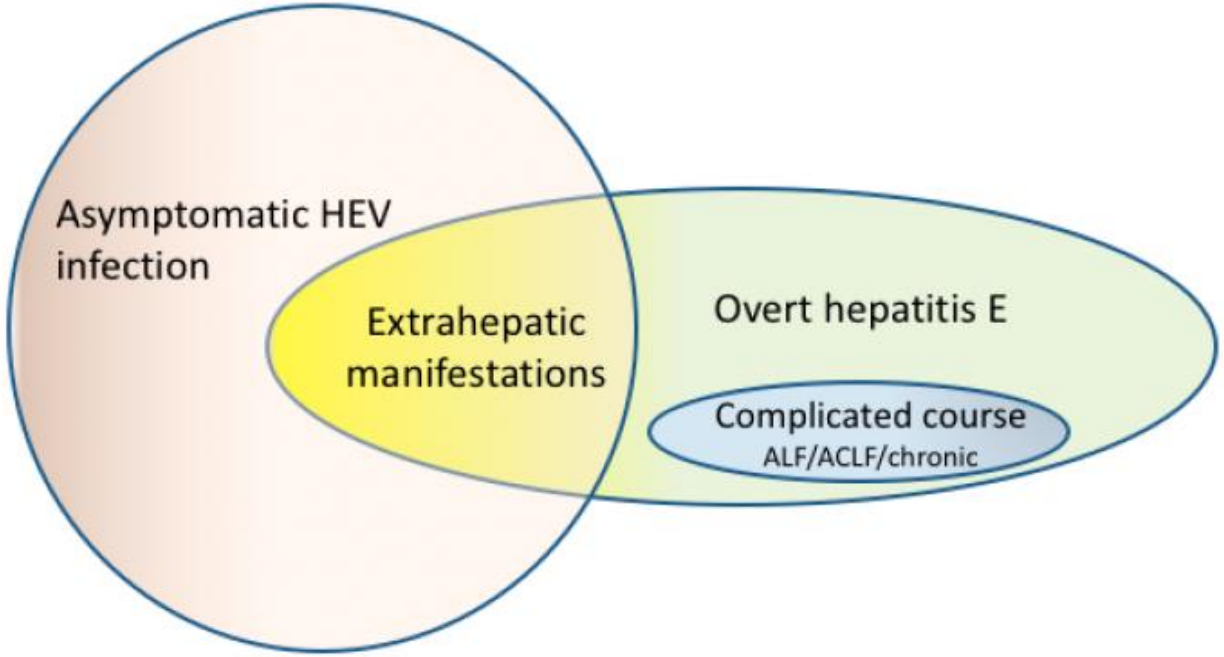


FIG 3 Course of acute hepatitis E virus (HEV) infection. Acute hepatitis E is characterized by symptoms such as fever, anorexia, vomiting, and jaundice, with onset several weeks after initial exposure. The onset of clinical symptoms coincides with a sharp rise in serum alanine transaminase (ALT) levels. Symptoms may persist for a few weeks to a month or more. ALT levels return to normal during convalescence. HEV RNA may be detected in both serum and stool early in the course of infection, but serum viremia may be difficult to detect by the time cases come to clinical attention. Anti-HEV IgM titers increase rapidly and then wane over the weeks following infection, while anti-HEV IgG antibody titers continue to rise more gradually during the convalescent period, and detectable anti-HEV IgG may persist for months to years. (Adapted from Fig. 2 of reference 170, with permission of the publisher [copyright 2012 Massachusetts Medical Society].)

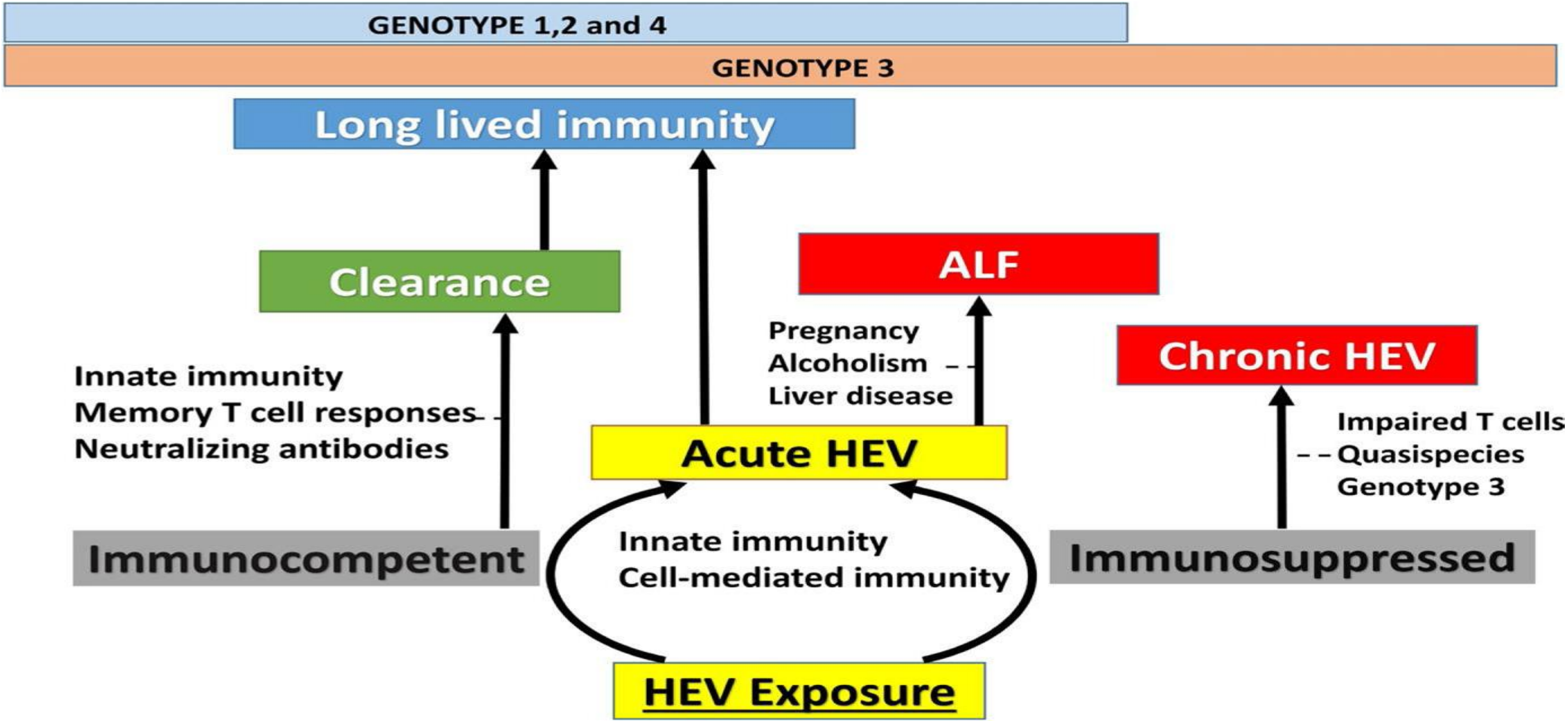
Sintomi

- **L'AHE è una malattia autolimitante con un basso tasso di mortalità.**
- **Aree endemiche l'infezione sintomatica è più comune a 15-40 anni; nei bambini asintomatica o lieve senza ittero.**
- **Segni e sintomi di AHE: febbre lieve, anoressia, nausea e vomito per alcuni giorni; dolore addominale, prurito, eruzione cutanea o dolori articolari; ittero urine scure e feci pallide; e epatomegalia.**

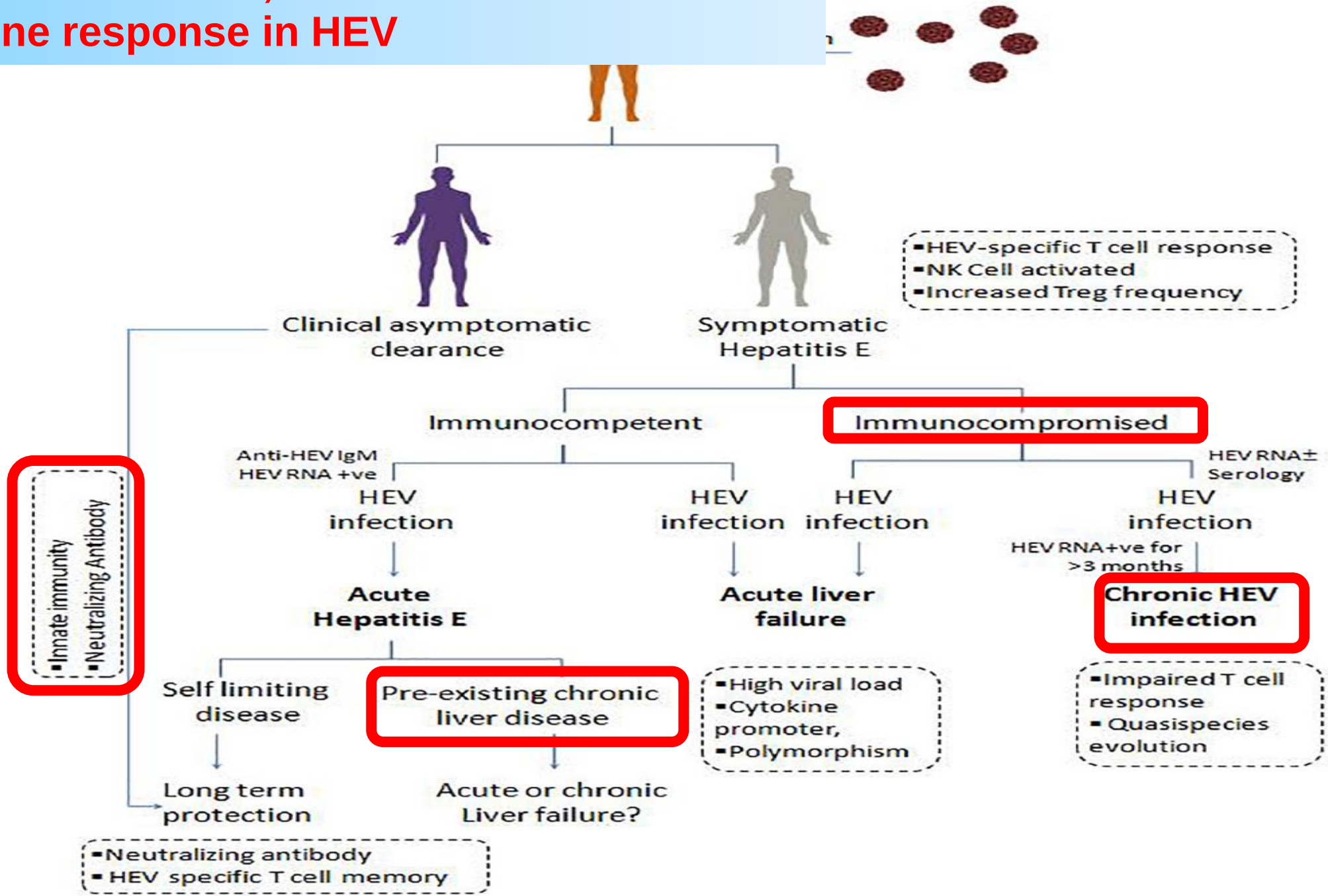
Nei pazienti immunocompromessi (SOT HEV3 o 4) presenza di HEV RNA per > 3 mesi si traduce in CHE (evoluzione HEV-quasispecie e dalla compromissione della risposta delle cellule T).



Natural course of HEV and its correlation with immune status and response. HEV 1, 2 and 4 cause acute self-limited disease as well as fulminant disease in immunocompetent host. HEV3 causes both acute disease as other genotypes and CHE in immunocompromised host.



Clinical manifestations, and immune response in HEV



HEV for High-Risk Patients

- **CHE is mainly seen in immunocompromised patients. Ankcorn et al. investigated CHE-patients lover long period of time and SOT was present in 60% of the patients.**
- **In 2 of 3 immunocompromised SOT-recipients, AHE leads to CHE, and in 10% to liver cirrhosis.**

TABLE 3 Prevalence of anti-HEV IgG and HEV RNA among transplant patients

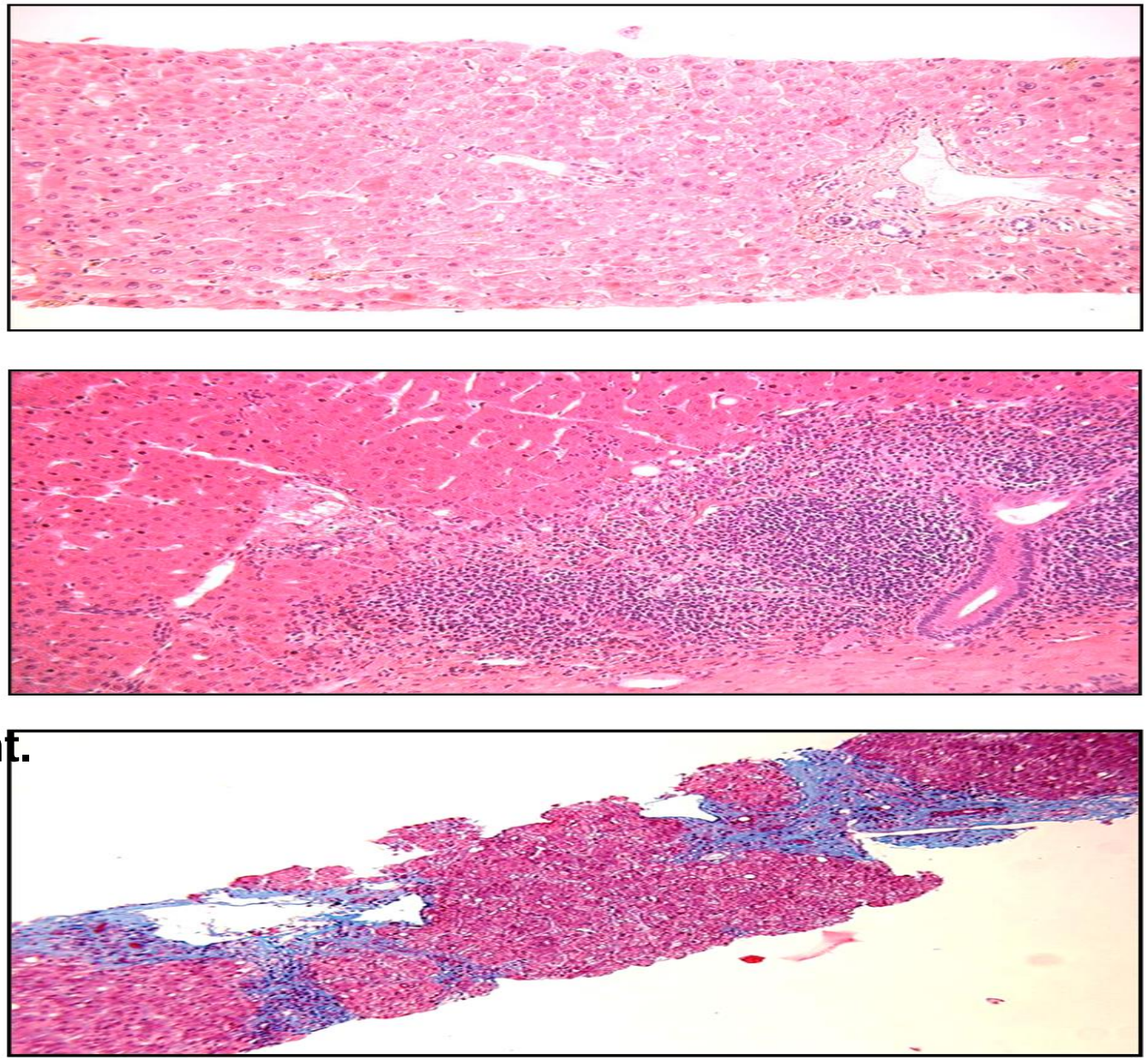
Study	No. of patients	Transplanted organ(s)[a]	Serology test	Prevalence (%)	
				Anti-HEV IgG	HEV RNA
Abravanel et al. (286)	88	SCT	Wantai	36	
Buti et al. (236)	108	KT, LT	Biokits	2.3	
Haagsma et al. (237)	285	LT	Genelabs	3.5	1.75
Kamar et al. (13)	217[b]	KT, LT, SPK	Abbott		6.5
Knoenecke et al. (285)	52	SCT	Abbott	5.3	0
Legrand-Abravanel et al. (188)	700	KT, LT, SPK	Adaltis	12.7	3.2
Moal et al. (238)	1,350	KT	Adaltis		1.2
Moal et al. (242)	160[b]	KT	Adaltis	14	4.3
Pas et al. (239)	1,200	SOT	Wantai		1
Pischke et al. (227)	274	HT	Genelabs	11.3	1.5
Pischke et al. (240)	226	LT	Abbott	4.4	0.9
Riezebos-Brilman et al. (241)	468	Lung transplantation			2.1

[a] SCT, stem cell transplantation; KT, kidney transplantation; LT, liver transplantation; SPK, simultaneous kidney-pancreas transplantation; SOT, solid organ transplantation; HT,

Nearly 10% of HEV-SOT patients develop cirrhosis within 3 to 5 years after primary infection

CHE have more aggressive variants (recombinant HEV-host variants).

PPR regions of recombinant variants included fragments of human genes of varying origin (inter alpha trypsin inhibitor (ITI-H2), ribosomal genes S17 or S19 and tyrosine aminotransferase). Variants harboring S17, S19 or ITI fragment had replicative advantage. Duplications and insertions of HEV genome were described.



Histology of liver biopsies from a CHE- patient.
(a) Initial liver biopsy,
(b) inflammation after 15 months of infection,
(c) Cirrhosis after 38 months of infection
(Masson's trichrome, magnification 100).

CHE in SOT

- L'epatite fulminante HEV non è riportata nei pazienti sottoposti a trapianto.
- HEV-Incidenza in SOT: 0,9-3,5%, (HEVRNA pos); in SOT-LT patients is 20%.
- > 60% dei pazienti con HEV abbia sviluppato CHE in trapianto di cuore .
- AHE diventano CHE in 21- 60%.
- L'attivazione della via di segnalazione IFN non porta alla clearance di HEV. L'aumento dell'espressione degli ISG in CHE-patients favorisce la persistenza del virus, la via di segnalazione IFN diventi refrattaria.
- I pazienti immunocompetenti non producono una tale risposta immunitaria, suggerendo che i pazienti SOT mobilitano una frazione maggiore della loro immunità a causa del trattamento farmacologico immunosoppressivo.

- Strong therapeutical immunosuppression to prevent rejection, SOT-pts can develop opportunistic infections.
- Risk factors in development of CHE-SOT: low platelet count, choice of immunosuppressant, liver cirrhosis and HIV infection.

SOT: Il regime immunosoppressivo può anche influenzare lo sviluppo di un'infezione cronica

- **HEV3-Suini trattati con ciclosporina, azatioprina e prednisolone hanno sviluppato CHE.**
- **HEV ha maggiori probabilità di persistere nei SOT trattati con tacrolimus rispetto a ciclosporina.**
- **Tacrolimus: fattore di rischio per il suo effetto immunosoppressivo più elevato rispetto alla ciclosporina A.**

Solo l'acido micofenolico (MFA) era associato alla clearance HEV. MFA deve essere presa in considerazione in HEV-SOT

Rapamicina ed everolimus promuovono la replicazione dell'HEV.

Immunocompromised SOT-recipients

The first treatment of SOT is reduction of immunosuppressive (to clear HEV in 30%)

Balance between infection and rejection (elevated risk of organ rejection)

- **The standard of care for patients unable to clear HEV is RBV 3-month.**
- **RBV for 3 months in 59 CHE-SOT recipients decrease HEV-replication (SVR 78%). Relapse patients retreated with RBV for 6 months cleared HEV with SVR 85%.**
- **CHE-LT and hemodialysis pts with Peg-INF-alfa 3 month results in HEV clearance (risk acute rejection). INF is contraindicated in kidney, pancreas, heart, lung transplant recipients because it increases risk of acute rejection.**
- **In vitro, sofosbuvir inhibits HEV replication and additive effect combination with RBV. CHE treated with sofosbuvir monotherapy showed decreased HEV replications but did not achieve viral clearances.**
- **In CHE adding sofosbuvir to RBV led to temporary HEV eradication.**

Patients with Hematological Diseases

- **L'insufficienza immunitaria dovuta a malattia di base o CT, il trapianto di cellule staminali è stato implicato come causa di HEV prolungato.**
- **HEV è associato a NHL. La prevalenza di HEV è più alta nei pazienti con malattia ematologica maligna rispetto alla popolazione generale (decorsi gravi o prolungati 28% della malattia HEV).**
- **Durante rituximab è stato osservato aumento delle transaminasi e dei marcatori HEV riducendo il dosaggio di rituximab da solo.**
- **È preferibile la somministrazione precoce di RBV, poiché la riduzione dell'immunosoppressione è stata associata ad un aumento della mortalità.**
-

Biol Blood Marrow Transplant 26 (2020) 1355–1362

Biology of Blood and Marrow Transplantation

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Hepatitis E Virus Infection in an Italian Cohort of Hematopoietic Stem Cell Transplantation Recipients: Seroprevalence and Infection

Elisa Furfaro[1], Laura Nicolini[2], Andrea Della Vecchia[1,2], Carmen Di Grazia[3], Anna Maria Raiola[3], Riccardo Varaldo[3], Fabio Ferrando[4], Gaia Barisione[1], Bianca Bruzzone[5], Emanuele Angelucci[3], Claudio Viscoli[1,2], Malgorzata Mikulska[1,2,*]

Aim: Italian cohort of 596 HSCT recipients analyze risk factors for HEV seropositivity (retrospective study, 2010-2019).

- Ha incluso pazienti sottoposti a trapianto 2010-2015 per i quali erano disponibili campioni di siero pre-trapianto (n = 419) e post-trapianto (n = 161) e testati retrospettivamente, nonché pazienti in cui è stato eseguito test prospettico HEV durante la cura standard: screening IgG pre-HSCT in 144, screening pre-HSCT HEV-RNA oltre allo screening IgG in 60 e test HEV-RNA in caso di sospetto clinico di infezione da HEV in 59.
-

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Table 1
Patient Characteristics

Characteristic	All Patients* (N = 596)	Patients Who Underwent HEV IgG Screening at HSCT (N = 563)
Age at HSCT, yr, median (range)	49 (17-74)	48 (17-74)
Male sex, n (%)	334 (56)	318 (56.5)
Underlying disease, n (%)		
Acute myeloproliferative disorders (AML/MDS)	256 (42.9)	237 (42.1)
Acute lymphoproliferative disorders (ALL and aggressive lymphomas)	139 (23.3)	133 (23.6)
Chronic lymphoproliferative disorders (CLL, MM, indolent lymphomas)	105 (17.6)	99 (17.6)
Chronic myeloproliferative disorders (CML and MF)	66 (11.1)	65 (11.5)
Severe aplastic anemia	28 (4.7)	27 (4.8)
Multiple sclerosis	2 (.3)	2 (.4)
Type of donor, n (%)		
Autologous	45 (7.5)	43 (7.6)
Allogeneic		
Matched related	150 (25.2)	142 (25.2)
Matched unrelated	63 (10.6)	60 (10.7)
Haploidentical	332 (55.7)	312 (55.4)
Cord blood	6 (1)	6 (1.1)

AML indicates acute myelogenous leukemia; MDS, myelodysplastic syndrome; ALL, acute lymphoblastic leukemia; NHL, non-Hodgkin lymphoma; CLL, chronic lymphoid leukemia; MM, multiple myeloma; CML, chronic lymphoid leukemia; SAA, severe aplastic anemia; MS, multiple sclerosis.

* Including those who underwent HEV IgG screening and an additional 33 tested with HEV-RNA because of clinical suspicion of HEV infection.

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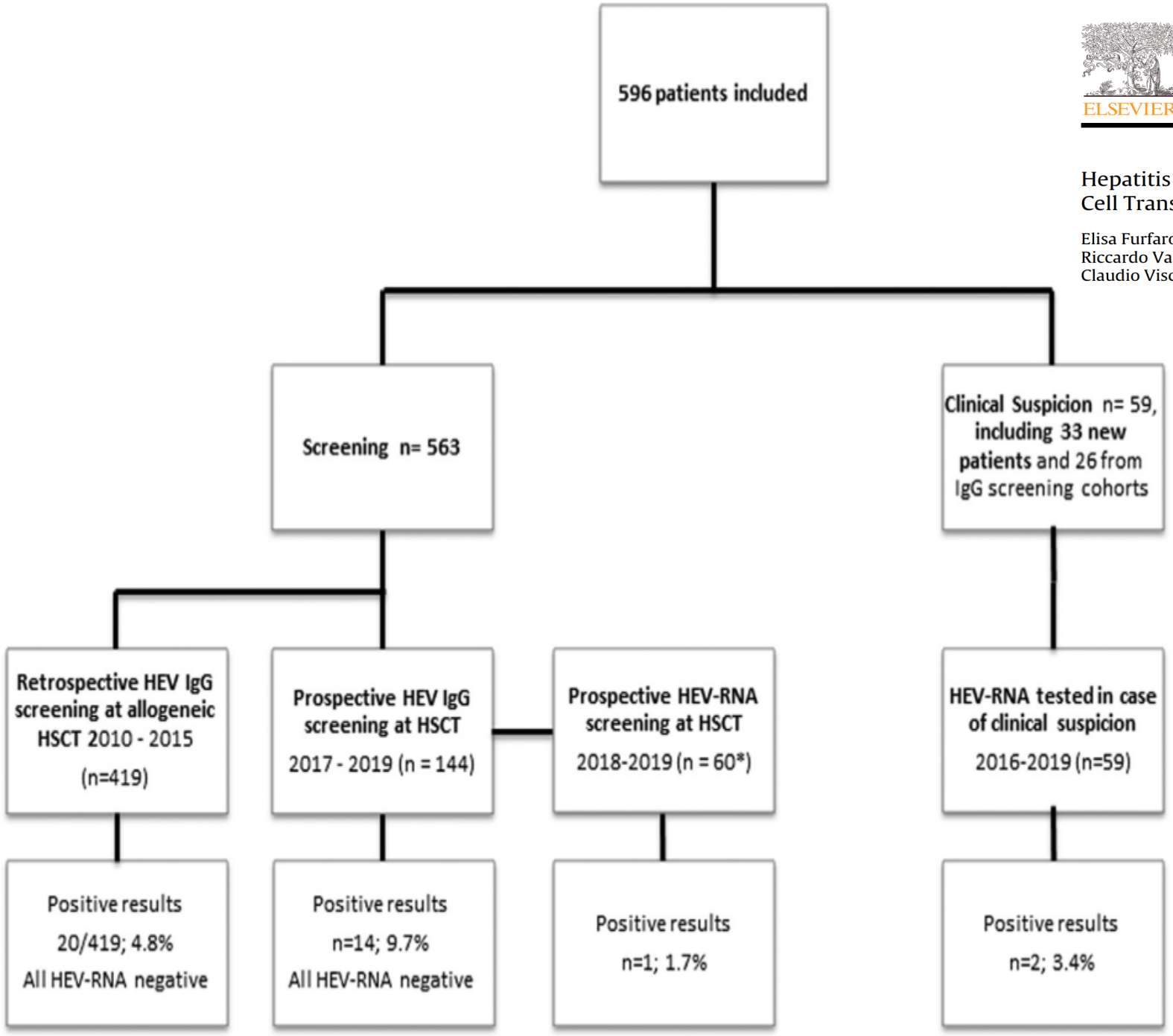

None of the 34 HEV-IgG-positive patients had detectable HEV-RNA. One case of transient HEV-RNA positivity pre-HSCT was identified through screening.

2 patients were diagnosed with chronic HEV hepatitis, and 1 patient was successfully treated with ribavirin.

Table 3
Univariate and Multivariate Analysis of Risk Factors for Positive HEV IgG Serology in 563 Patients from 2010 to 2019

Risk F				
Age, y				
Sex, n				
Mal				
Fem				
Year o				
Under				
Acu				
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Allo				
HEV Ig				
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HEV				

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* Con

Rate of pre-HSCT HEV-IgG positivity: 6.0% (34 of 563).
Older age: an independent risk factor for seropositivity (P = .039).

None of the 34 HEV-IgG-positive patients had detectable HEV-RNA.
One case of transient HEV-RNA positivity pre-HSCT was identified through screening.

2 patients were diagnosed with chronic HEV hepatitis, and 1 patient was successfully treated with ribavirin.

The burden of HEV infection in HSCT recipients in Italy is limited, and pre-HSCT screening appears to be of no benefit.

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Ulteriori gruppi di pazienti immunocompromessi

- **Più del 10% di CHE.**
- **L'infezione da HIV CD4+ inferiore a 200 cellule/mm3 è il principale fattore di rischio per CHE. Trattare con RBV, peg-INF alfa, o una combinazione di entrambi. La normalizzazione della conta dei CD4+ da sola non porta alla risoluzione dell'HEV stesso.**

Malattie reumatiche Il metotrexato ha molteplici effetti sulle cellule T. Alcuni pazienti che ricevono metotrexato raggiunto HEV-SVR dopo la riduzione dell'immunosoppressione.

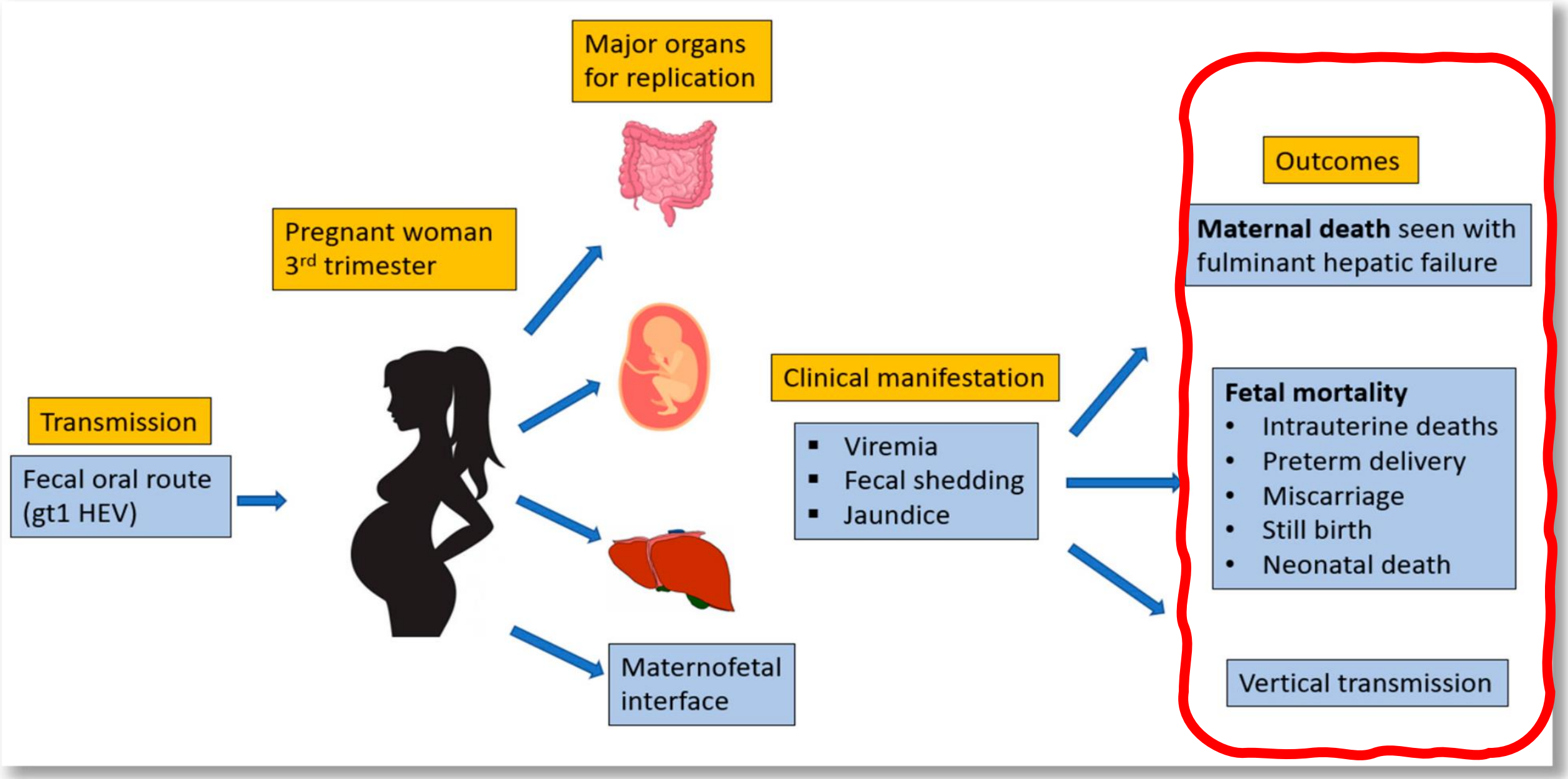
Gli inibitori del TNF CHE richiede la riduzione della terapia. La produzione di citochine pro-infiammatorie come TNF svolge un

Severe Hepatitis in Pregnant Women (PW)

HEV-PW nel 2 o 3 trimestre hanno aumentato rischio di insufficienza epatica acuta, perdita fetale e mortalità. 20-30% mortalità se HEV contratto nel 3 trimestre nei paesi in via di sviluppo. HEV1 si replica in placenta con gravi danni ai tessuti (> apoptosi e necrosi all'interfaccia materno-fetale con alterazioni della barriera placentare) rispetto a HEV3. HEV1 produce più virioni infettivi e innesca produzione di citochine pro-infiammatorie (IL-6, chemochine)).

Table 1. HEV outbreaks (serologically confirmed) in pregnant women in different countries.

No.	Country	Year	Total cases	Deaths	References
1	Nepal	1987	73	18	54
2	India	1987	19	3	55
3	India	1988	30	2	56
4	Somalia	1988–1989	346	48	57
5	Indonesia	1991	19	5	58
6	Kenya	1991	28	4	59
7	India	1991	48	12	24
8	Djibouti	1991–1993	3	1	60
9	Pakistan	1993–1994	35	4	61
10	Central African Republic	2002	7	1	62
11	Sudan	2004	61	19	63
12	Uganda	2007–2009	189	13	64
13	Bangladesh	2008–2009	21	4	65
14	Bangladesh	2010	12	3	66
15	Sudan	2010–2011	39	11	67



- **Non sono disponibili regimi terapeutici stabiliti per le donne in gravidanza. (RBV teratogeno)**

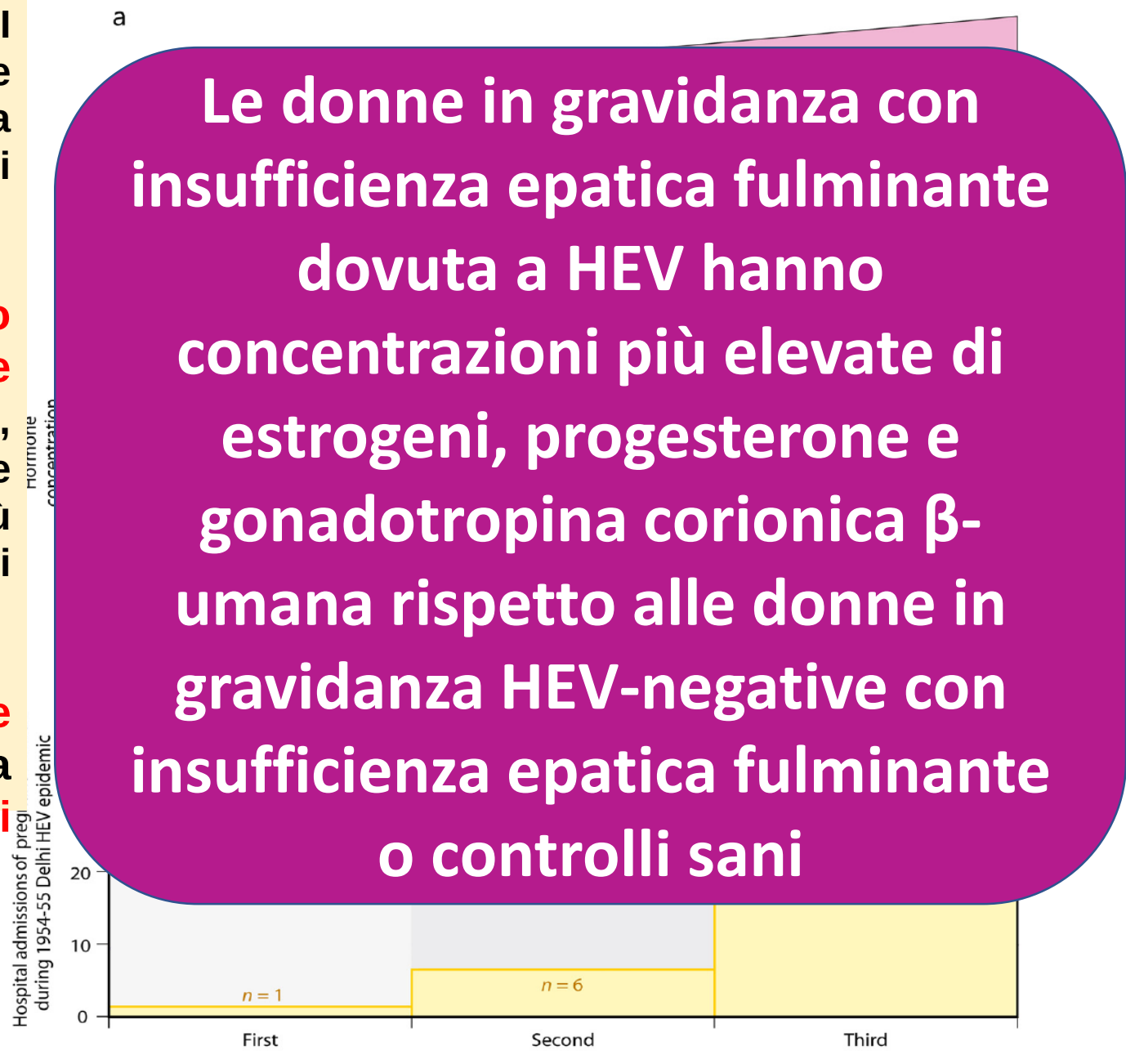
PW: Con il progredire della gravidanza, RI adattativa diminuisce e il numero e funzione delle cellule T e NK diminuiscono costantemente. La RI si allontana dalla linea Th1, diminuiscono le cellule B. barriera immunitaria è rafforzata dall'aumento fagocitosi e dall' attività dei granulociti.

Le alterazioni degli ormoni svolgono ruolo dominante. Fino 3 trimestre, > estrogeno e progesterone. All'inizio della gravidanza, estrogeno > risposta Th1 e immunità dipendente dalle cellule. Con concentrazioni di estrogeni più elevate, risposta Th2 ha maggiori probabilità di essere supportata insieme alla risposta umorale.

Alte concentrazioni di progesterone sopprimono la RI materna e l'interazione tra Th1 e Th2. livelli elevati di estrogeni promuovono HEV-replicazione.

Portano al parto pretermine e mortalità fetale nei pazienti con HEV per la disfunzione placentare

Le donne in gravidanza con insufficienza epatica fulminante dovuta a HEV hanno concentrazioni più elevate di estrogeni, progesterone e gonadotropina corionica β-umana rispetto alle donne in gravidanza HEV-negative con insufficienza epatica fulminante o controlli sani



In PIU’ HEV induce…

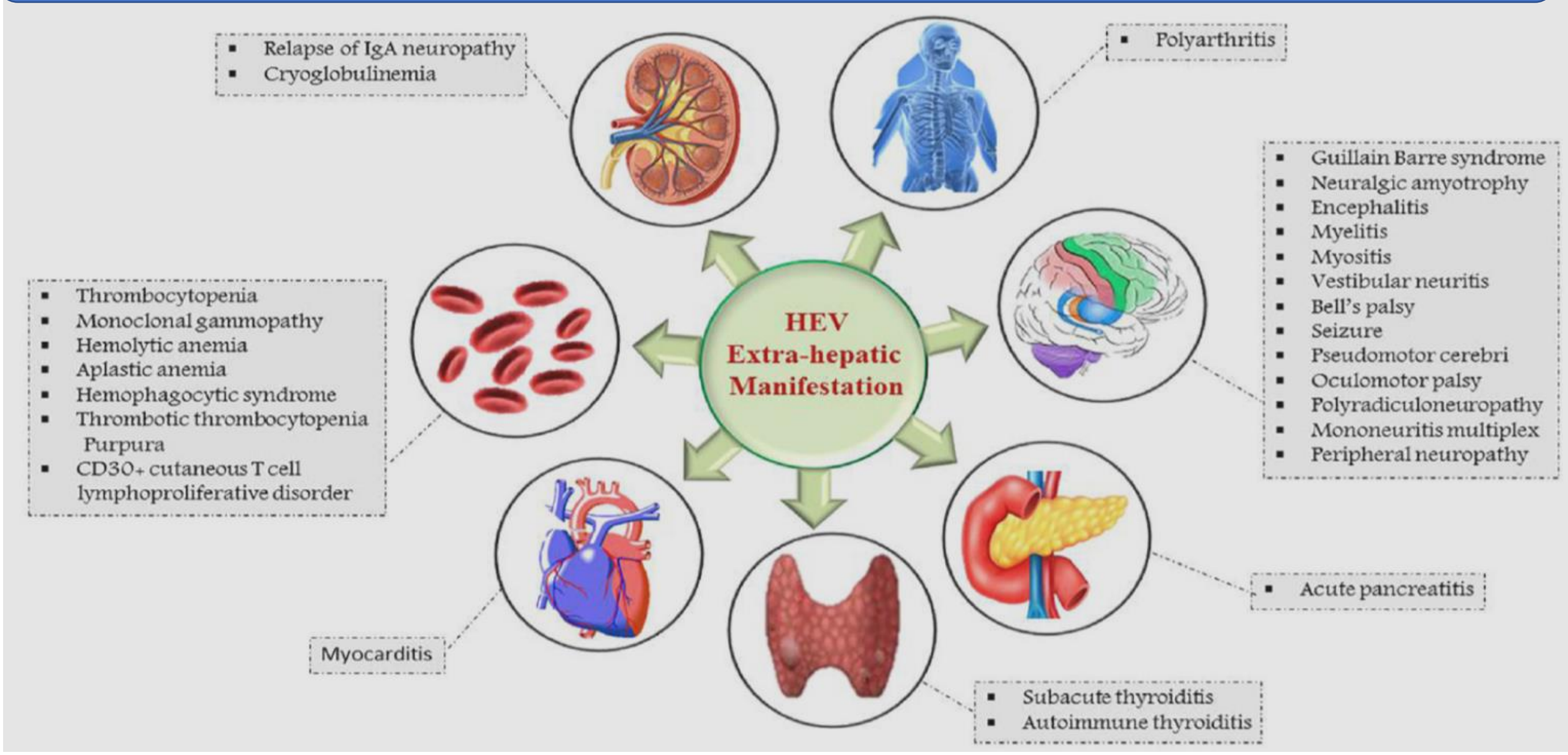
HEV1 prolifera in modo più efficiente di HEV3 ex vivo nelle cellule stromali e negli espianti di tessuto di decidua e placenta fetale.

Cambiala struttura della barriera placentare con aumento della morte cellulare e necrosi all'interfaccia materno-fetale nelle donne in gravidanza con infezione da HEV1.

HEV1 porta alla produzione di virioni più infettivi e citochine pro-infiammatorie come le chemochine e IL-6.

Questi cambiamenti nel microambiente delle citochine sono associati a elevata carica virale e aumento del danno tissutale

Le reazioni immuno-mediate e il danno tissutale HEV diretto (citopatico) sono i meccanismi colpevoli nelle manifestazioni extraepatiche



Manifestazioni extraepatiche

Replicazione diretta dell'HEV nei tessuti extraepatici, dimostrata nella placenta e nel liquido cerebrospinale.

manifestazioni ematologiche: anemia emolitica, anemia aplastica e trombocitopenia grave. 12/106 (11%) pazienti con HEV avevano una bassa conta piastrinica.

pancreatite acuta in India Il 2,1% (16/790) dei pazienti aveva HEV-1 recente (edema ampolla di Vater).

manifestazioni autoimmuni: miocardite, artrite e tiroidite.

Manifestazioni neurologiche

- **Il danno al sistema nervoso periferico è predominante.**
- **Lesioni neurologiche: amiotrofia nevralgica (NA), sindrome di Guillain-Barré, paralisi di Bell e poliradicolopatia, dolore neuropatico, disturbi sensoriali indolori, encefalite, mielite, miosite, mononeurite multipla, neurite vestibolare, neurite brachiale bilaterale, neuropatia periferica.**
- **studio prospettico europeo (Francia, Regno Unito e Paesi Bassi) su 450 pazienti consecutivi con lesioni neurologiche non traumatiche ad esordio acuto: 2,4% HEV. I 3 casi di NA erano associati a HEV.**
- **Studio coorte anglo/olandese: 5/47 (10,6%) pazienti con NA avevano HEV all'inizio della malattia.**
- **studio europeo multicentrico su 118 pazienti con NA ha dimostrato che pazienti con HEV hanno fenotipo clinico distinto (più coinvolgimento bilaterale e danni più estesi al plesso brachiale) rispetto a quelli senza HEV.**
- **Dimostrata HEV RNA e sintesi intratecale di anti-HEV IgM in pazienti con NA.**

Manifestazioni neurologiche

- **L'associazione tra HEV e sindrome di Guillain-Barré è supportata da 3 studi caso-controllo in Bangladesh, Giappone e Paesi Bassi. Hanno trovato HEV recente nel 5-11% dei pazienti con sindrome di G-B.**

  6/73 (8%) pazienti belgi con sindrome di G-B avevano infezione da HEV.

NA si verificano dopo l'infezione da HEV. I meccanismi fisiopatologici??? Le lesioni neurologiche sono più frequenti nei pazienti immunocompetenti (22,6%) rispetto a quelli immunocompromessi (3,2%, p < 0,001), suggerendo che sindrome di G-B e NA sono immuno-mediate a causa del mimetismo molecolare.

Neurotropismo virale diretto: sindrome piramidale è stata osservata nel ricevente del rene CHE. Le varianti caratterizzate nel liquido cerebrospinale di questo paziente sono diverse da quelle nel siero, suggerendo l'emergere di varianti neurotropiche e la replicazione dell'HEV nel SNC.

Quasi tutti questi pazienti con manifestazioni neurologiche avevano una funzionalità epatica normale o modestamente anormale, indicando che i sintomi e i segni neurologici dominano il quadro clinico in questi

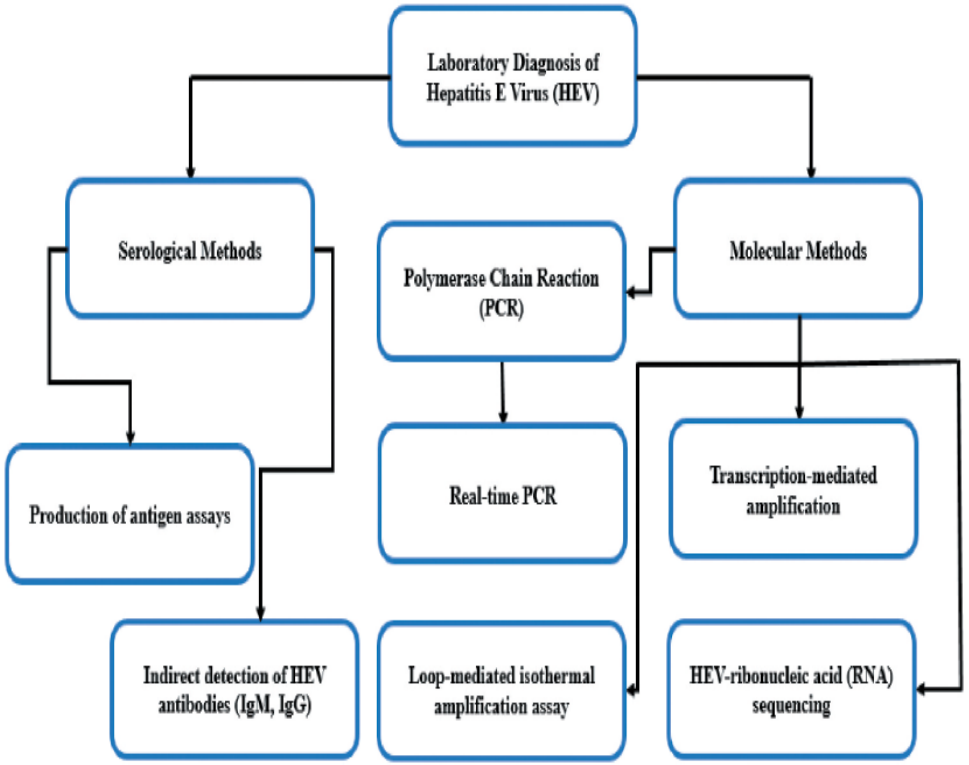
Manifestazioni renali

- **HEV può portare a lesioni renali e compromissione della funzionalità renale, si verificano dopo l'infezione da HEV-1,3 (6%).**
- **HEV: fattore predittivo indipendente per crioglobulinemia nei riceventi SOT, l'infezione da HEV provoca diminuzione dell'eGFR. La funzionalità renale migliora dopo clearance dell'HEV e la proteinuria si riduce. Glomerulonefrite associata alla crioglobulina è probabilmente innescata dall'esuberante risposta immunitaria agli antigeni virali.**
- **Nessuna prova che l'HEV sia direttamente nefrotossico o che replica nelle cellule renali.**
- **App. Urinario serbatoio HEV: alte concentrazioni di Ag HEV e nelle urine di pazienti immunocompromessi. HEV-Ag potrebbe essere secreto nelle urine dalle cellule epiteliali renali.**
- **Biopsie renali in HEV1,3: malattia glomerulare (glomerulonefrite membranoproliferativa (MPGN) con/senza crioglobululinemia e glomerulonefrite membranosa) HEV può innescare riacutizzazioni di nefropatia IgA pre-esistente. L'HEV RNA è stato rilevato nel crioprecipitato dal siero di**

Conclusions

- The pathogenesis of HEV infection involves a complex interplay between the virus and the host immune response.
- While the clinical phenotype of an HEV infection is primarily hepatological, the range and incidence of HEV-associated clinical symptoms is much broader, including neurological and renal disorders.
- Ribavirin is currently the treatment of choice for chronically infected patients but there is an urgent need to find other compounds with which to treat patients who fail to clear HEV after ribavirin therapy.
- MORE SCREENING

Diagnosi AHE: IgM anti-HEV nel siero e/o HEV RNA nel siero o feci. Genoma HEV: PCR generica o RT-PCR.Metodi basati su Ab (Western blot o immunoassay enzimatici) CHE: IgG anti-HEV
Infettività di HEV è determinata nell'inoculazione in animali o con coltura cellulare



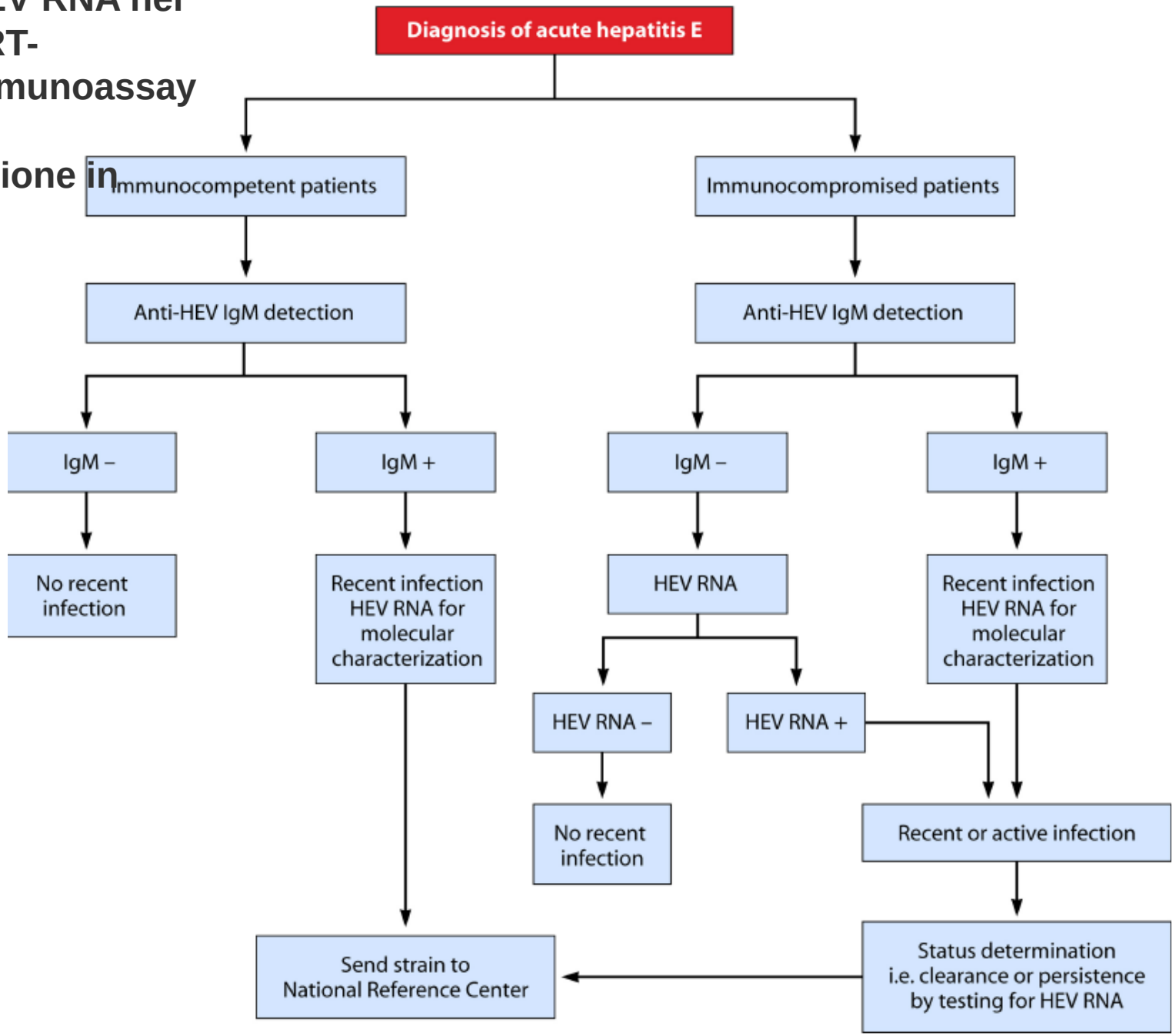
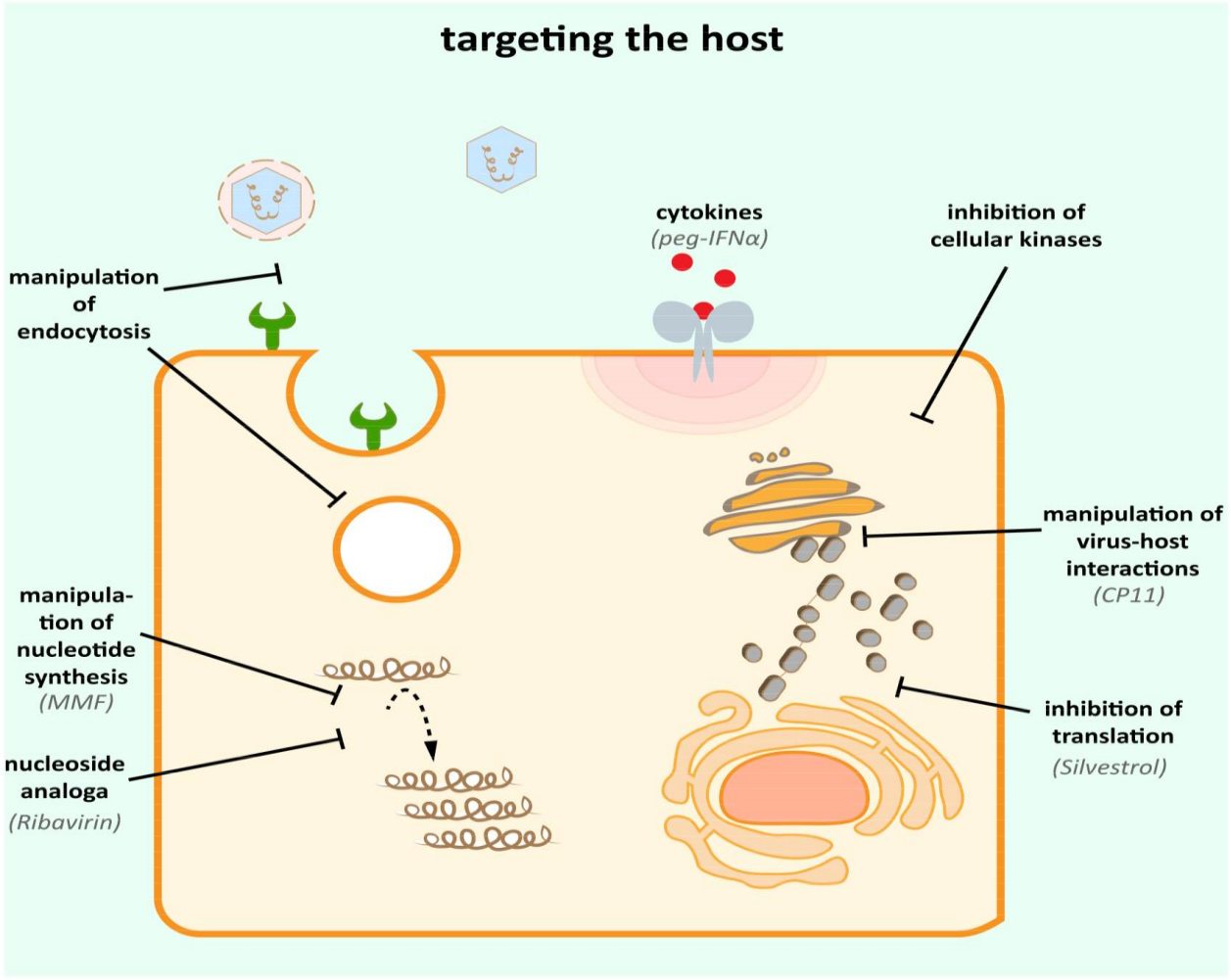
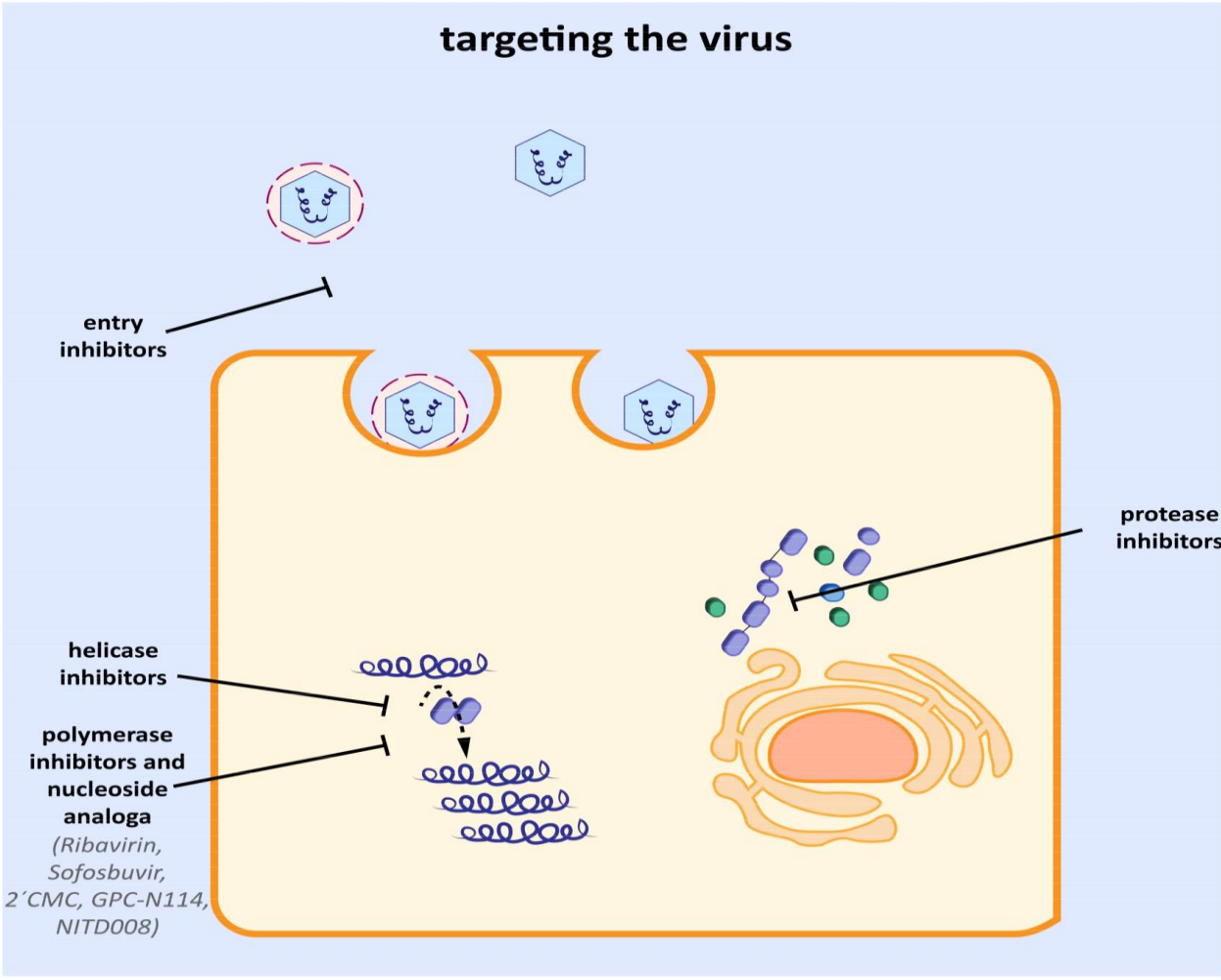


FIG 4 Flow diagram for diagnosis of acute hepatitis E virus infection

Potential host and viral targets of antiviral drugs

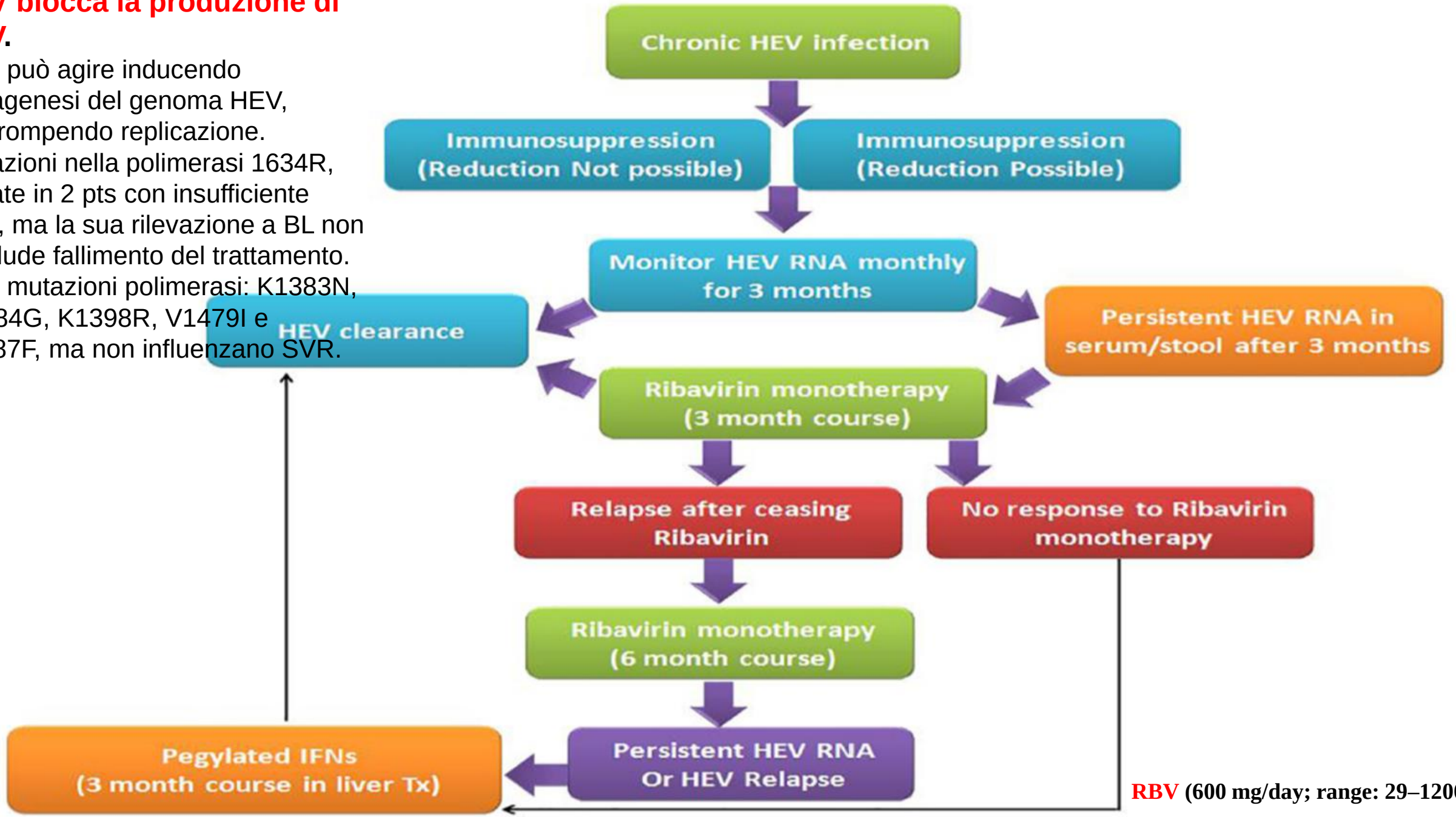




Antiviral therapy against HEV can rely on inhibition and manipulation of host components, which are important for HEV life cycle. Direct acting antiviral specifically can target viral enzymes (helicase, polymerase, protease) without affecting host components. RBV is reported to exert antiviral effects by targeting both virus and host

RBV blocca la produzione di HEV.

RBV può agire inducendo mutagenesi del genoma HEV, interrompendo replicazione.
Mutazioni nella polimerasi 1634R, trovate in 2 pts con insufficiente RBV, ma la sua rilevazione a BL non preclude fallimento del trattamento.
HEV mutazioni polimerasi: K1383N, D1384G, K1398R, V1479I e Y1587F, ma non influenzano SVR.



RBV (600 mg/day; range: 29–1200)

Immunocompromised SOT-recipients

The first treatment of SOT is reduction of immunosuppressive (to clear HEV in 30%)

Balance between infection and rejection (elevated risk of organ rejection)

- **The standard of care for patients unable to clear HEV is RBV 3-month.**
- **RBV for 3 months in 59 CHE-SOT recipients decrease HEV-replication (SVR 78%). Relapse patients retreated with RBV for 6 months cleared HEV with SVR 85%.**
- **CHE-LT and hemodialysis pts with Peg-INF-alfa 3 month results in HEV clearance (risk acute rejection). INF is contraindicated in kidney, pancreas, heart, lung transplant recipients because it increases risk of acute rejection.**
- **In vitro, sofosbuvir inhibits HEV replication and additive effect combination with RBV. CHE treated with sofosbuvir monotherapy showed decreased HEV replications but did not achieve viral clearances.**
- **In CHE adding sofosbuvir to RBV led to temporary HEV eradication.**

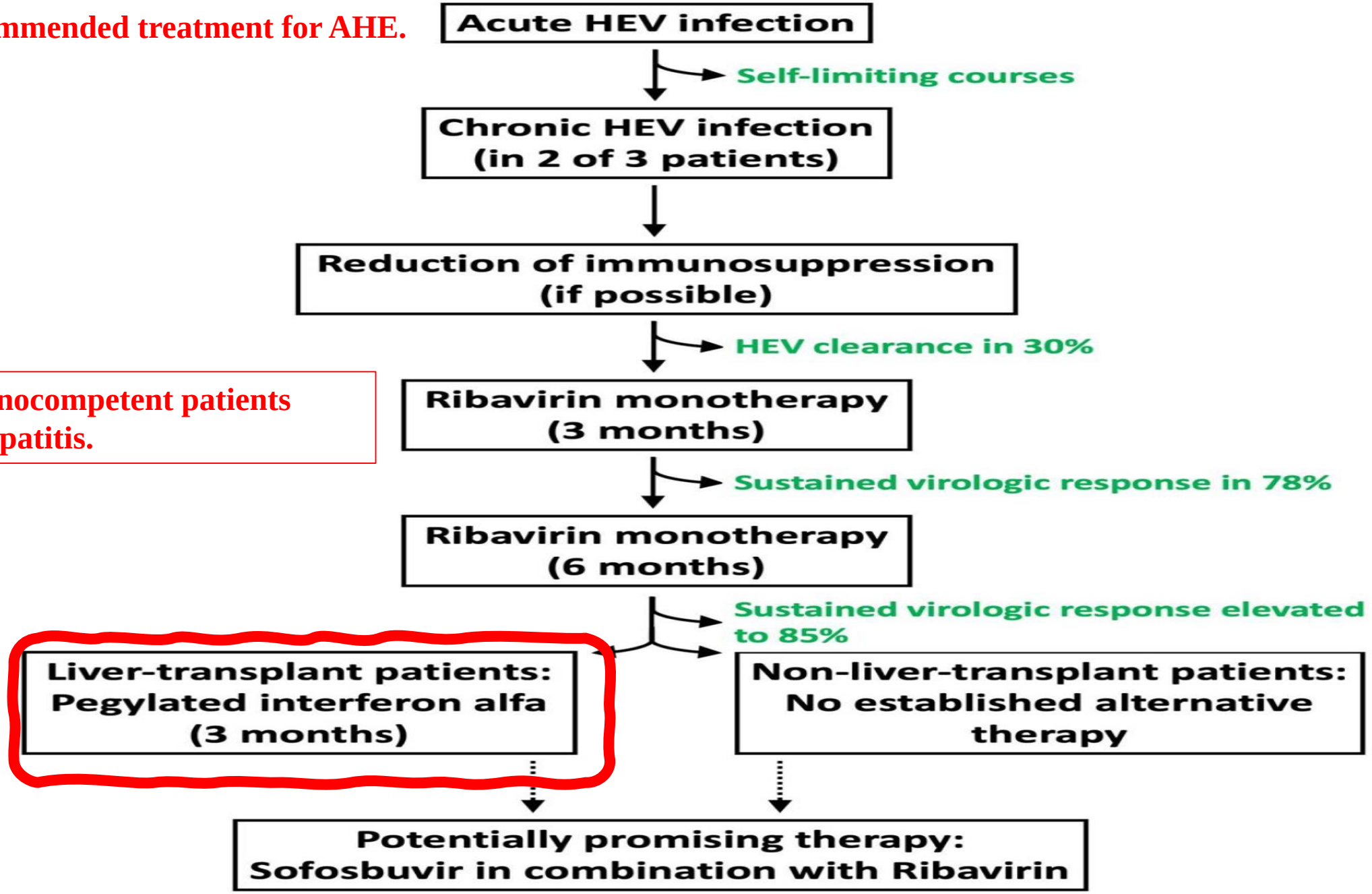
Treatment

- **Sofosbuvir was shown to have antiviral activity against HEV in vitro but had limited efficacy in HEV-patients.**

A pilot study of 9 patients (7 RBV previously failed) given sofosbuvir (400 mg/day) for 24 wks found that their HEV viremia decreased, but was not cleared.

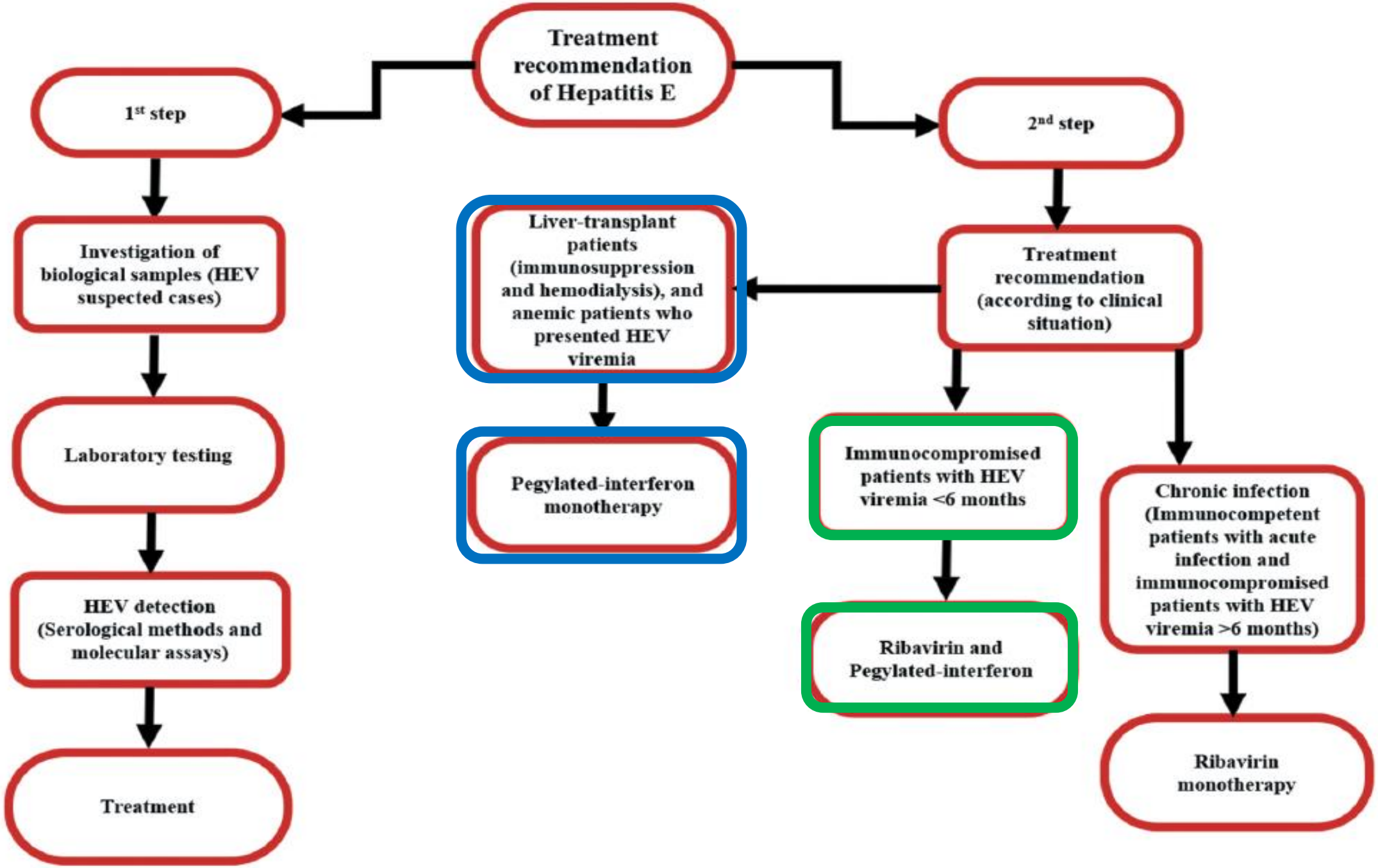
- **Sofosbuvir alone is not cure for HEV.**

There is no recommended treatment for AHE.



RBV in immunocompetent patients with severe hepatitis.

Figure 2. Therapy algorithm for chronic HEV-infected immunosuppressed patients.


Treatment recommendation of Hepatitis E
1st step
Investigation of biological samples (HEV suspected cases)
Laboratory testing
HEV detection (Serological methods and molecular assays)
Treatment
2nd step
Treatment recommendation (according to clinical situation)
Liver-transplant patients (immunosuppression and hemodialysis), and anemic patients who presented HEV viremia
Pegylated-interferon monotherapy
Immunocompromised patients with HEV viremia <6 months
Ribavirin and Pegylated-interferon
Chronic infection (Immunocompetent patients with acute infection and immunocompromised patients with HEV viremia >6 months)
Ribavirin monotherapy

Ulteriori gruppi di pazienti immunocompromessi

- **Più del 10% di CHE.**
- L'infezione da HIV CD4+ inferiore a 200 cellule/mm3 è il principale fattore di rischio per CHE. Trattare con RBV, peg-INF alfa, o una combinazione di entrambi. La normalizzazione della conta dei CD4+ da sola non porta alla risoluzione dell'HEV stesso.
- Malattie reumatiche Il metotrexato ha molteplici effetti sulle cellule T. Alcuni pazienti che ricevono metotrexato raggiunto HEV-SVR dopo la riduzione dell'immunosoppressione.
- Gli inibitori del TNF CHE richiede la riduzione della terapia. La produzione di citochine pro-infiammatorie come il TNF svolge un ruolo nel controllo delle infezioni da HEV.

Patients with Hematological Diseases

- **Immune insufficiency due to underlying disease or CT, stem cell transplantation has been implicated as a cause of prolonged HEV.**
- **HEV is mainly associated with NHL. HEV prevalence is higher in patients with malignant hematological disease than in the general population (severe or 28% prolonged HEV disease courses).**
- **During rituximab was observed rising transaminases and HEV markers by reducing rituximab dosing alone.**
- **Early RBV administration is preferable, as reduction of immunosuppression was associated with increased mortality.**

Screening to identify new anti-HEV drugs: NITD008, broad-spectrum chain-terminating adenosine nucleoside analogue (dengue), GPC-N114, which binds to RNA channels of picornavirus polymerases.

T cell-based immunotherapy based on cytotoxic T cells targeting specific antigens. Soon et al. identified several HEV-specific T cell receptors that could be a potential candidates in therapy of CHE.

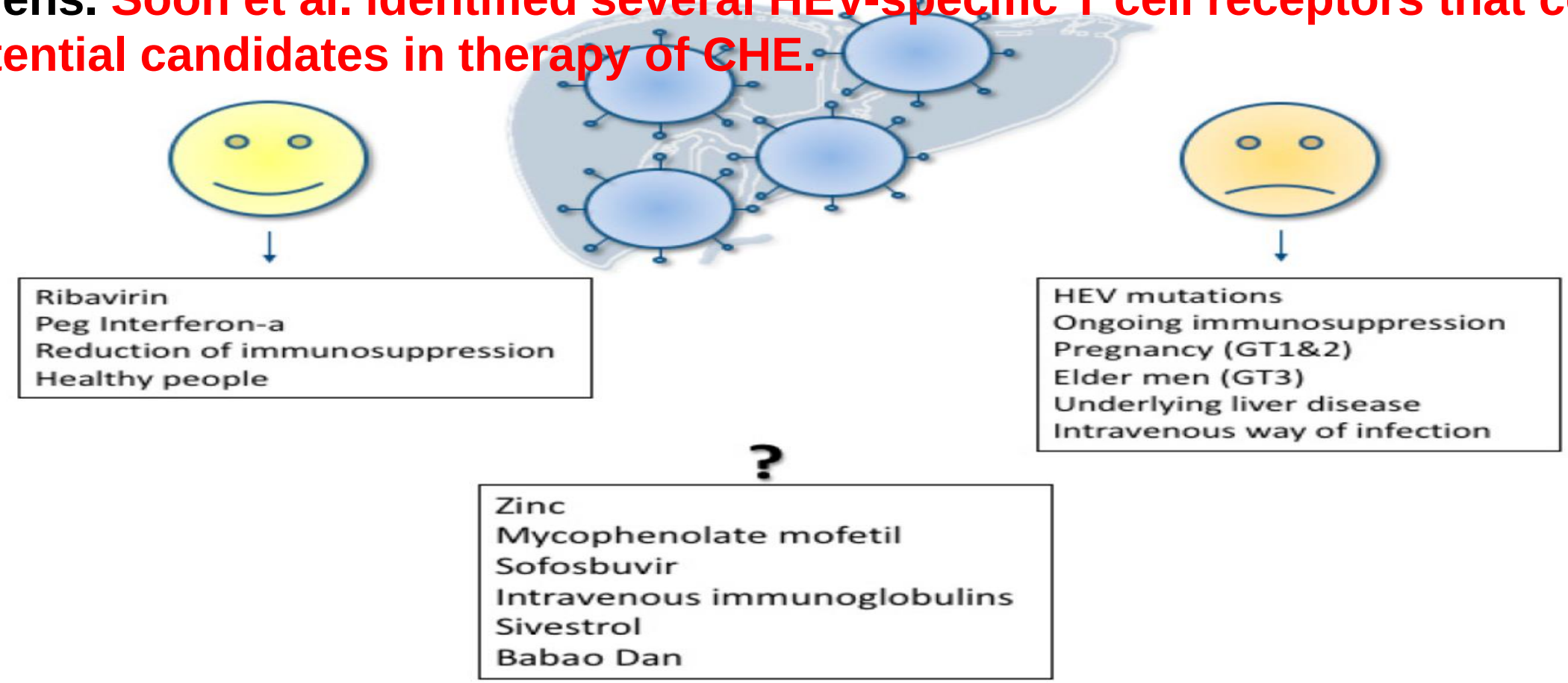
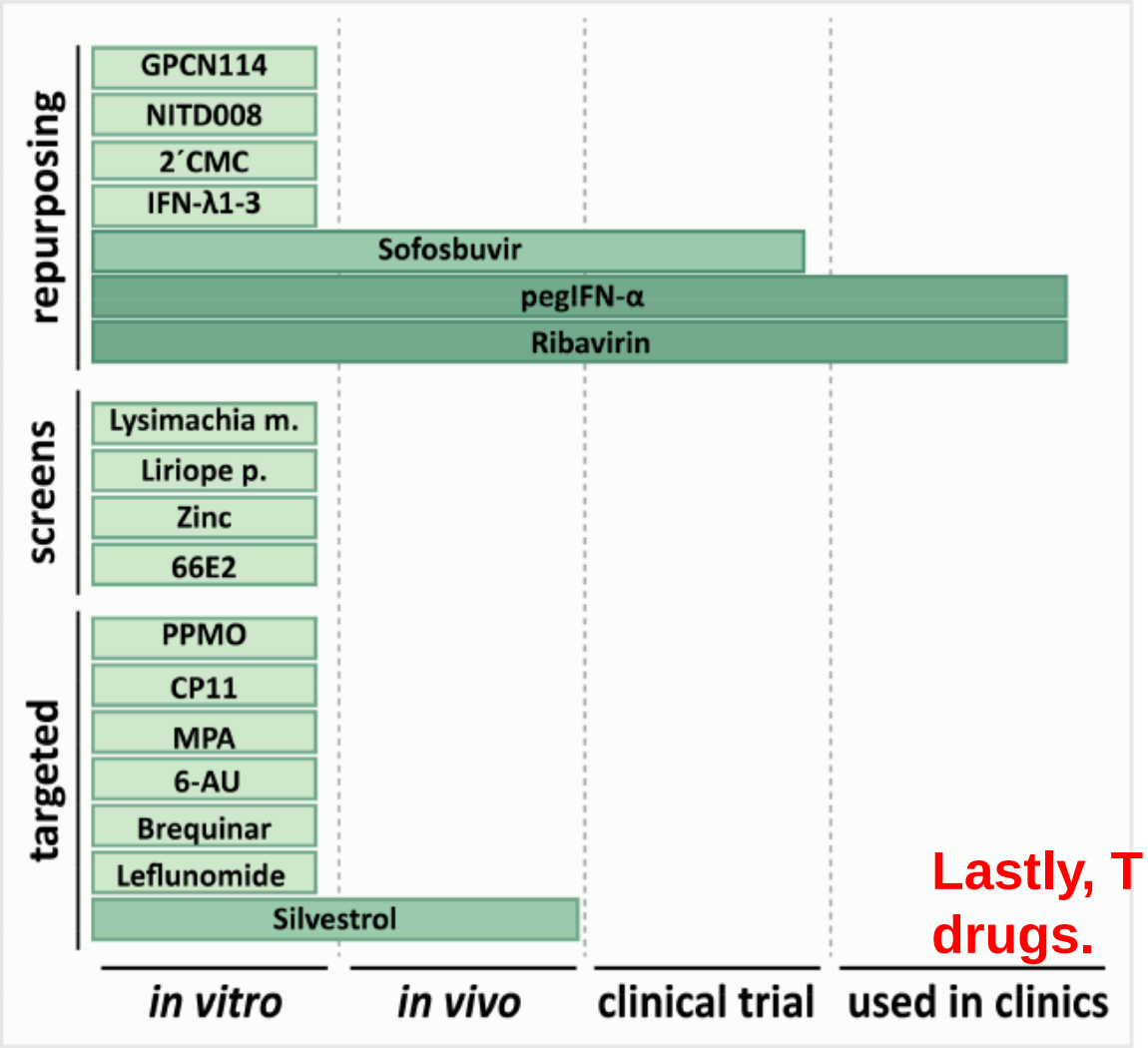


Figure 2. Different factors influencing HEV infection: Protective and beneficial factors on the left hand; unfavorable and risk factors on the right hand; not yet clarified factors in the lower middle.



The risk of HEV reinfection is unclear

Although minimum protective concentration of Ab remains to be determined, an Ab concentration of 2.5 WHO units/mL was found to be protective. In vaccine study SOT recipients can be re-infected when Ab concentration is below 7 WHO units/mL

Lastly, T cell therapy may be an alternative to drugs.

Figure 3. Overview of molecules/extracts with antiviral activity against HEV. The depicted molecules/extracts are classified according to the strategy that was used to identify them. So far, the antiviral activity against HEV of only four drugs (Sofosbuvir, pegIFN-α, Ribavirin and silvestrol) was approved in experimental settings beyond in vitro cell culture systems. Both drugs in the clinics are used off-label and therefore have not been tested in clinical trials against HEV.

2’-C-Methylcytidine

2'-C-metilcitidina (2-CMC), NM107, analogo nucleosidico (NA) contro HEV in vitro, composto ha dimostrato effetto inibitorio contro la replicazione dell'HEV (IC50 di 22 M/L). Insieme a RBV ha moderato effetto antagonistico, suggerendo meccanismo convergente di inibizione. Nel trattamento dell'HCV, è stata riportata biodisponibilità orale insufficiente. Il profarmaco NM283 sviluppo è stato interrotto per effetti tossici avversi.

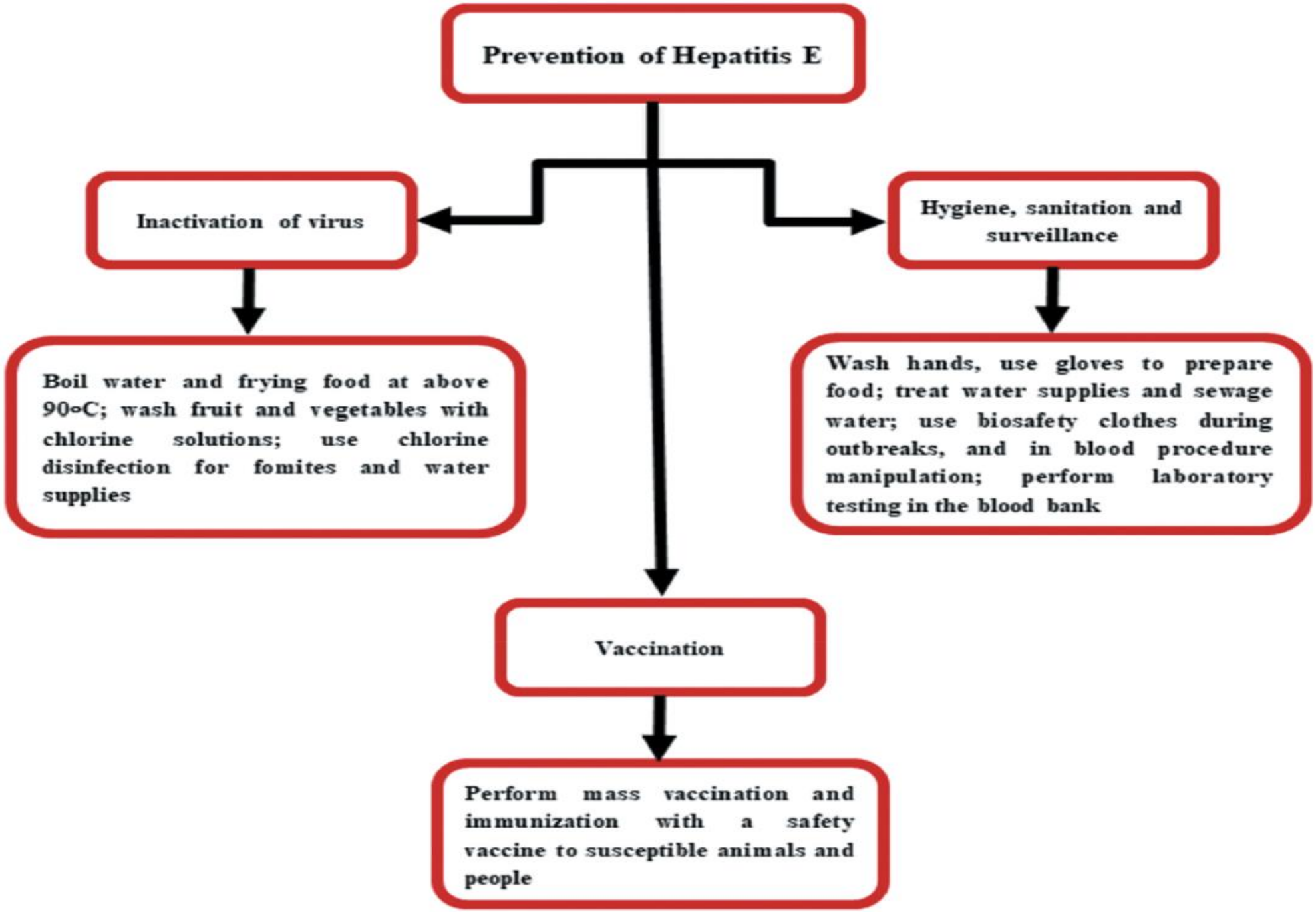
Zinc salts can block HEV replication in vitro by inhibiting RNA polymerase. 2 F CHE-transplant-recipients did not respond to RBV despite having elevated intra-erythrocyte zinc concentrations. The natural compound silvestrol blocks HEV replication in vitro, and HEV RNA concentrations in feces of treated mice rapidly decreased. has a similar action in humans …

NITD008 and GPC-N114

GPC-N114 e NITD008, inibendo replicone subgenomico derivato da GT1. GPC-N114 aveva concentrazione efficace semi-massima (EC50) di 1,07 M e indice terapeutico (TI) di >93 (GPC-N114) mentre NITD008 aveva EC50 0,03 M e TI >3333. GPC-N114 è stato descritto come NNI di RdRP di più generi all'interno della famiglia Picornaviridae. NITD008 agisce come antivirale su HCV, West Nile Virus o Zika. NITD008 ha mostrato tossicità in vivo.

Ciprofloxacin and IFN-

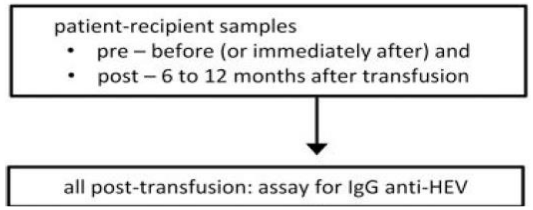
- Nishiyama et al hanno utilizzato farmaci per lo screening in vitro contro un ceppo GT3 contenente Gaussia luciferase reporter. Ciprofloxacina (CPFX) e IFN-1-3 hanno mostrato la migliore attività contro il genoma del reporter.
- Tutti i sottotipi IFN-, ma non CPFX, hanno mostrato una robusta riduzione dell'HEV RNA.


Prevention of Hepatitis E
Inactivation of virus
Boil water and frying food at above 90∘C; wash fruit and vegetables with chlorine solutions; use chlorine disinfection for fomites and water supplies
Hygiene, sanitation and surveillance
Wash hands, use gloves to prepare food; treat water supplies and sewage water; use biosafety clothes during outbreaks, and in blood procedure manipulation; perform laboratory testing in the blood bank
Vaccination
Perform mass vaccination and immunization with a safety vaccine to susceptible animals and people

Increasing the coverage of HEV vaccination will be key to minimizing the burden of AHE.

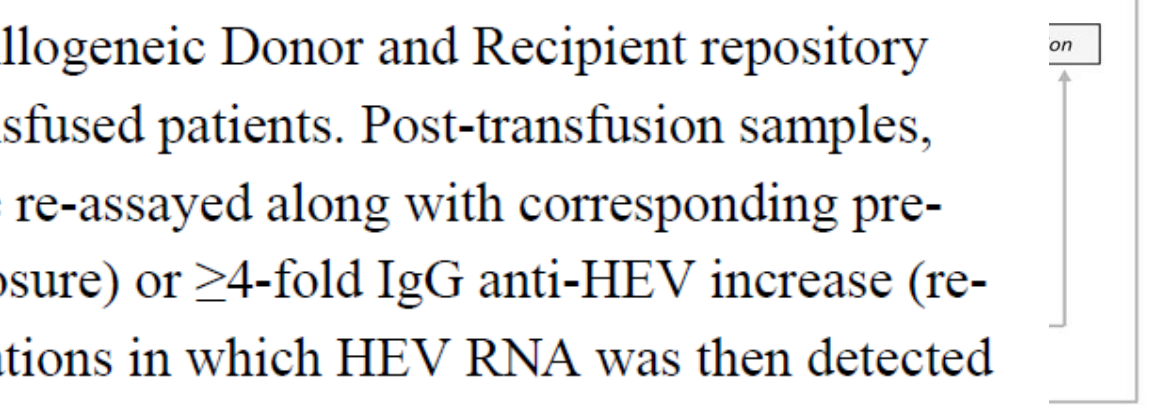
- (Hecolin®) vaccine developed in China elicits protective antibodies to all HEV genotypes (HEV1,4) and has been licensed in China since 2012 (Zhu 2010). This vaccine is 97% effective (3 dose course) in preventing symptomatic AHE (Zhang 2015).
- not been licensed and approved in any other country. It is not recommended by WHO in children aged under 16 years, PW, chronic liver disease, organ transplant waiting lists, and travellers .

Testing algorithm for patient-recipient and donation samples



- We HE cor sub yie dur ser HE pla tha cor ant ana det

STUDY DESIGN AND METHODS: To identify donations that contained HEV RNA and were linked to patient-recipients with antibody evidence of HEV exposure, we assayed samples from the Retrovirus Epidemiology Donor Study (REDS) Allogeneic Donor and Recipient repository that represents 13,201 linked donations and 3384 transfused patients. Post-transfusion samples, determined to contain IgG anti-HEV by ELISA, were re-assayed along with corresponding pre-transfusion samples for seroconversion (incident exposure) or ≥4-fold IgG anti-HEV increase (re-exposure). HEV-exposed patients were linked to donations in which HEV RNA was then detected by RT-qPCR, confirmed by Transcription Mediated Amplification (TMA), and phylogenetically analyzed as sub-genomic cDNA sequences.



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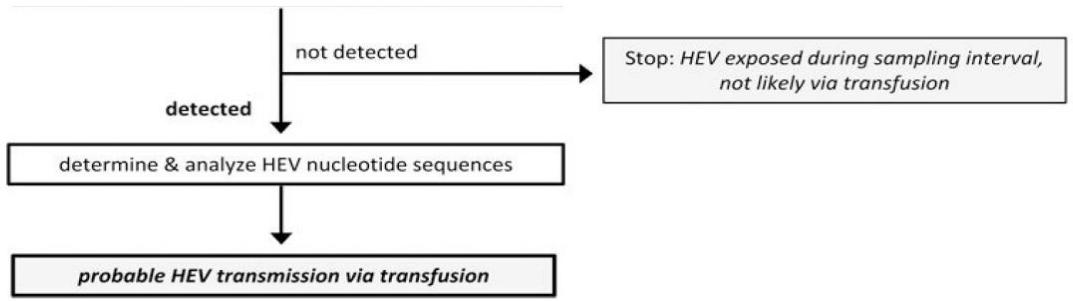
Probable Transmission of Hepatitis E Virus (HEV) via Transfusion in the United States

John R Ticehurst[1,4,6], Nora Pisanic[2], Michael S Forman[6], Carly Ordak[2], Christopher D Heaney[2,1,3], Edgar Ong[8,*], Jeffrey M Linnen[8,†], Paul M Ness[7,5], Nan Guo[1,‡], Hua Shan[9], and Kenrad E Nelson[1,4]



The risk of HEV reinfection is unclear.

Although the minimum protective concentration of antibodies remains to be determined, an antibody concentration of 2.5 World Health Organization (WHO) units/mL was found to be protective in a vaccine study SOT recipients can be re-infected when the antibody concentration is below 7 WHO units/mL

There is no recommended treatment for AHE.

- **The optimal duration of treatment has still to be determined.**
- **Treatment should be continued for further 3 months if HEV RNA is detected in stool at EOT.**

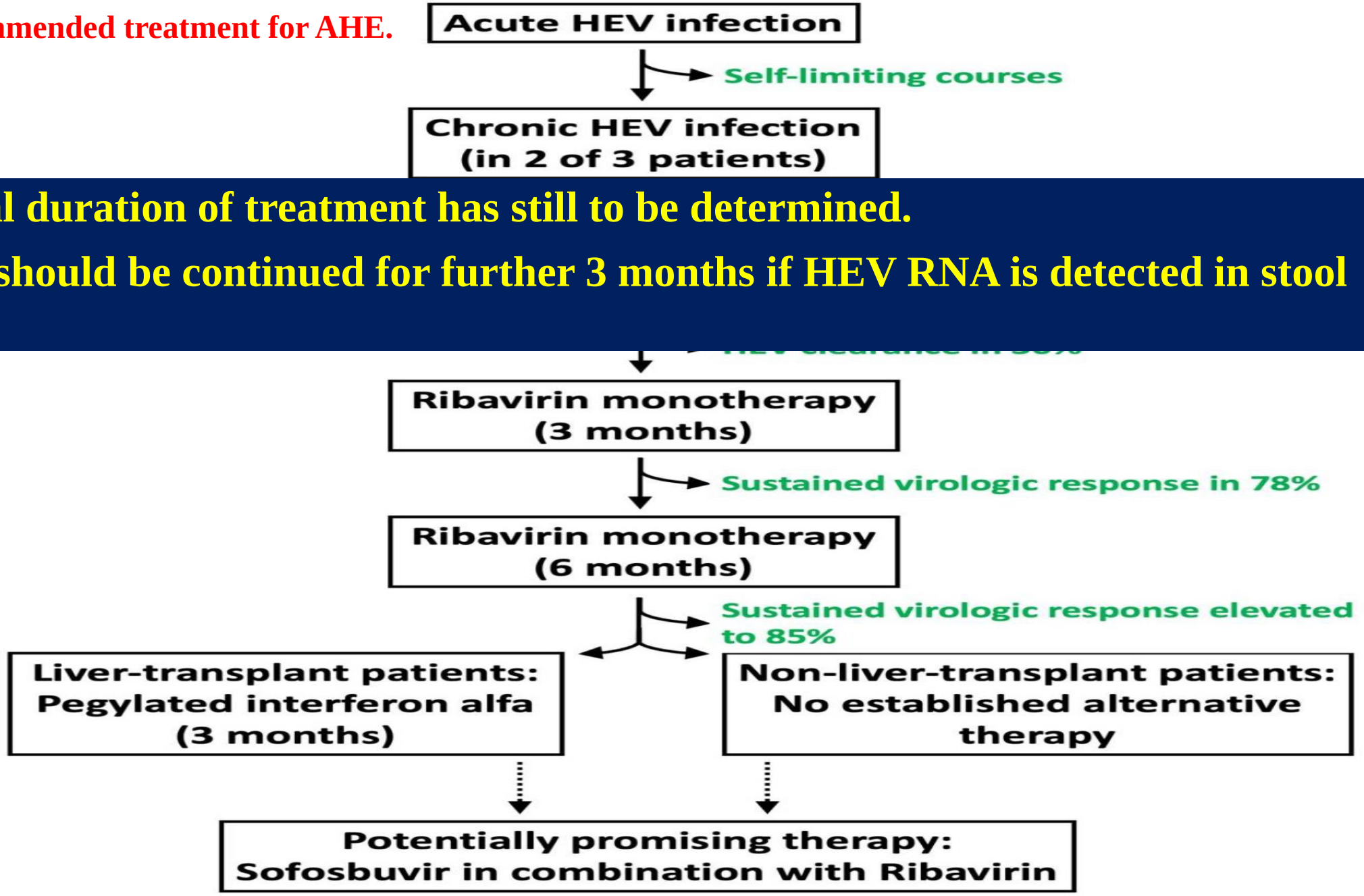


Figure 2. Therapy algorithm for chronic HEV-infected immunosuppressed patients.

Conclusions

- The pathogenesis of HEV infection involves a complex interplay between the virus and the host immune response. While the clinical phenotype of an HEV infection is primarily hepatological, the range and incidence of HEV-associated clinical symptoms is much broader, including neurological and renal disorders.
- Ribavirin is currently the treatment of choice for chronically infected patients but there is an urgent need to find other compounds with which to treat patients who fail to clear HEV after ribavirin therapy.

Table 1. Histopathological evaluation of liver tissue and results of IHC, serology and real-time PCR in HEV-positive wild boar.

Animal	Sex	Age	Real-time PCR HEV		Liver HEV viral load (copies/mL)	IHC HEV	Ab HEV	Hepatocellular necrosis	Lymphoplasmacytic aggregates	Sinusoidal congestion	Bile pigment accumulation
			Liver	Serum							
1	Female	Subadult	+	-	532	+	-	/	◆	▲	/
2	Female	Subadult	+	-	478	+	-	□	◆	▲	●
3	Female	Juvenile	+	-	107	+	-	/	◆	▲▲	/
4	Female	Adult	+	+	426	+	-	/	◆◆	▲▲▲	●

Real-time PCR hepatitis E virus (HEV), immunohistochemistry (IHC) HEV and antibodies (Ab) HEV: (-) negative; (+) positive.

Hepatocellular necrosis:(/) absence; (□) <10 cells per total section of 1cm^2 approx.

Lymphoplasmacytic aggregates: (/) absence; (◆) ≤3 small aggregates; (◆◆) 4–7 small or medium-sized aggregates.

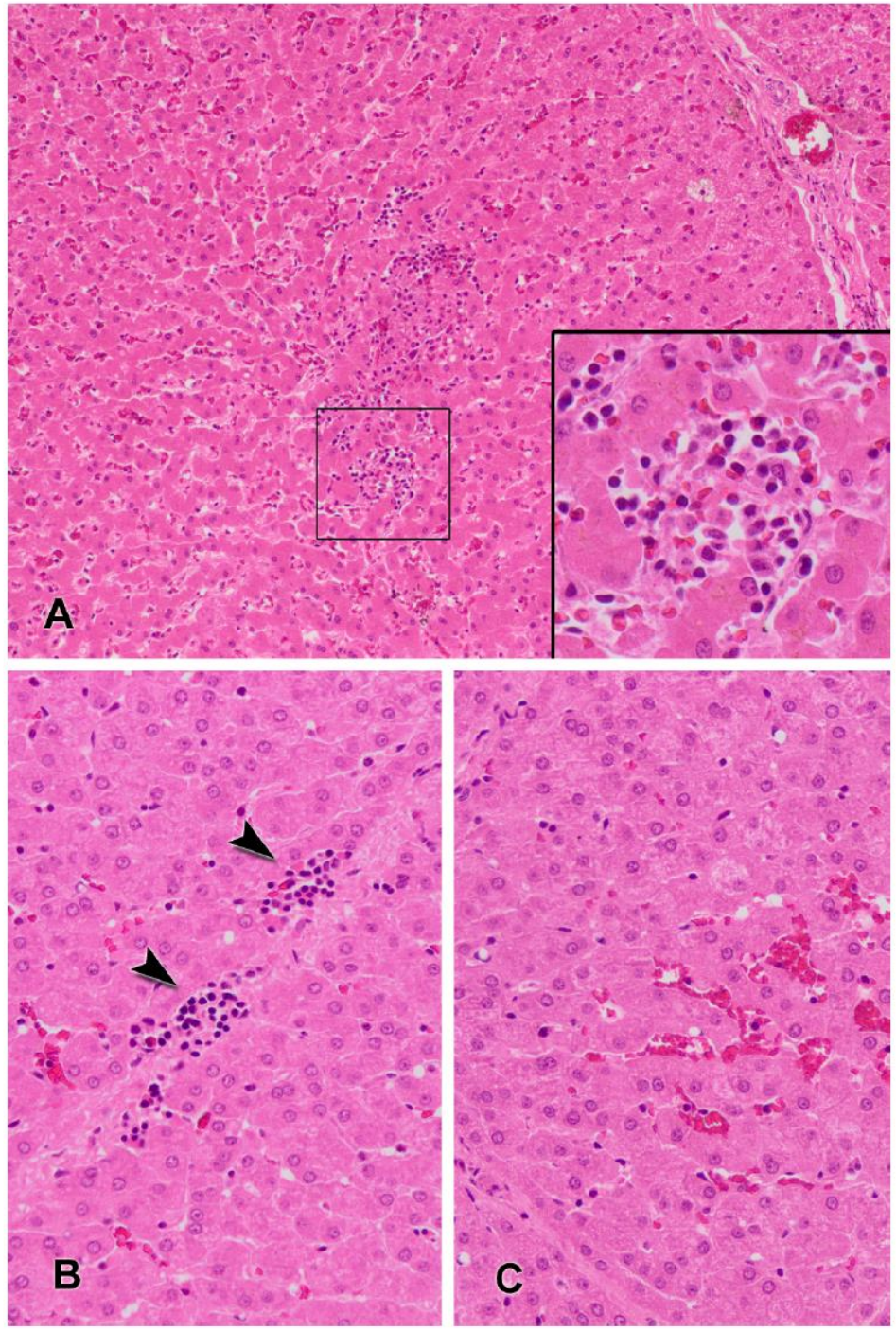
Sinusoidal congestion: (▲) very scarce presence of erythrocytes; (▲▲) ≈30% of sinusoids contain moderate amounts of erythrocytes; (▲▲▲) 30–80% of lobules are highly congested.

Bile pigment accumulation (extracellular (canalicular cholestasis) and intracellular): (/) absence; (●) multifocal and in low amounts.

Histopathologic lesions in wild boar naturally infected with HEV

- The livers of the 4 animals with naturally occurring HEV infection displayed only mild diffuse lymphoplasmacytic infiltrates (Fig 1A and 1B), with one of them also showing slight signs of hepatocyte necrosis. One of these animals also had a high degree of congestion in the hepatic sinusoids (Fig 1C).

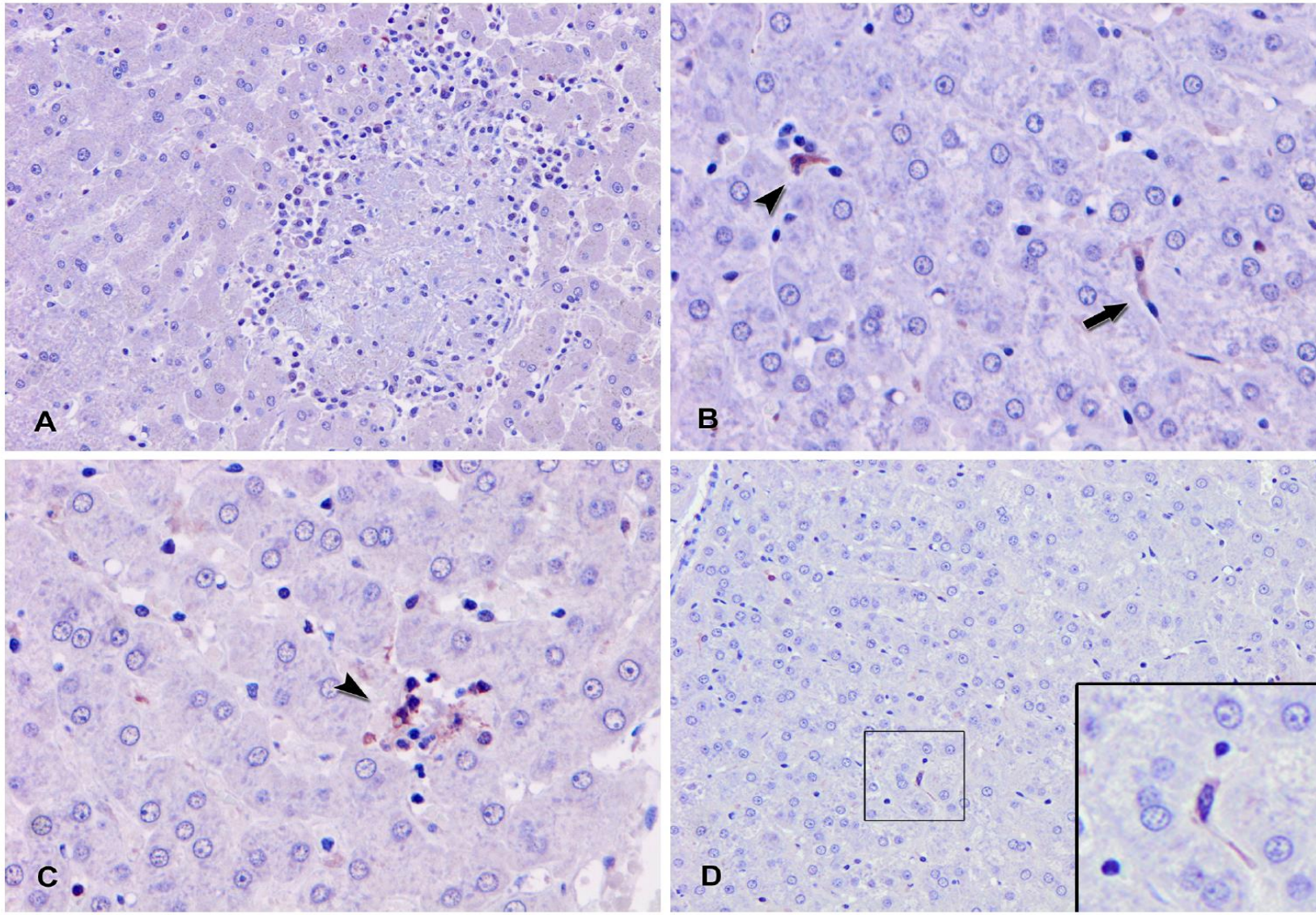
- Fig 1A: liver with presence of mild intralobular lymphoplasmacytic aggregates in the hepatic parenchyma. Inset showing a lymphoplasmacytic aggregate at higher magnification (hematoxylin and eosin, HE).
- Fig 1B: liver showing slight perilobular infiltrations of lymphocytes (arrowheads) (HE).
- Fig 1C: hepatic lobule with moderate
- hyperemia of liver sinusoids (HE).



PLOS ONE | https://doi.org/10.1371/journal.pone.0186858

Representative photomicrographs of liver sections from 3 wild boar naturally infected with HEV

- Fig 2A: liver with absence of association of HEV antigen inside lymphoplasmacytic aggregates (Immunohistochemistry (IHC) with avidin-biotin-peroxidase complex (ABC) method counterstained with hematoxylin).
- Fig 2B: HEV immunolabeling in cytoplasm of Kupffer cells (arrowhead) and sinusoidal endothelial cells (arrow) (IHC ABC method).
- Fig 2C: HEV immunostaining of light lymphoplasmacytic infiltrate (arrowhead) (IHC ABC method).
- Fig 2D: liver showing slight labeling for HEV antigen in hepatic parenchyma, which is appreciable in Kupffer cells (inset) (IHC ABC method).



PLOS ONE | https://doi.org/10.1371/journal.pone.0186858

2’-C-Methylcytidine

- La 2'-C-metilcitidina (2-CMC), NM107, analogo nucleosidico (NA) contro HEV in vitro, il composto ha dimostrato un effetto inibitorio contro la replicazione dell'HEV (IC50 di 22 M/L).
- Applicandolo insieme a RBV ha prodotto un moderato effetto antagonistico, suggerendo un meccanismo convergente di inibizione.
- Quando valutato per il trattamento dell'HCV, è stata riportata una biodisponibilità orale insuciente. è stato sviluppato un profarmaco NM283, lo sviluppo è stato interrotto a causa di effetti tossici avversi.
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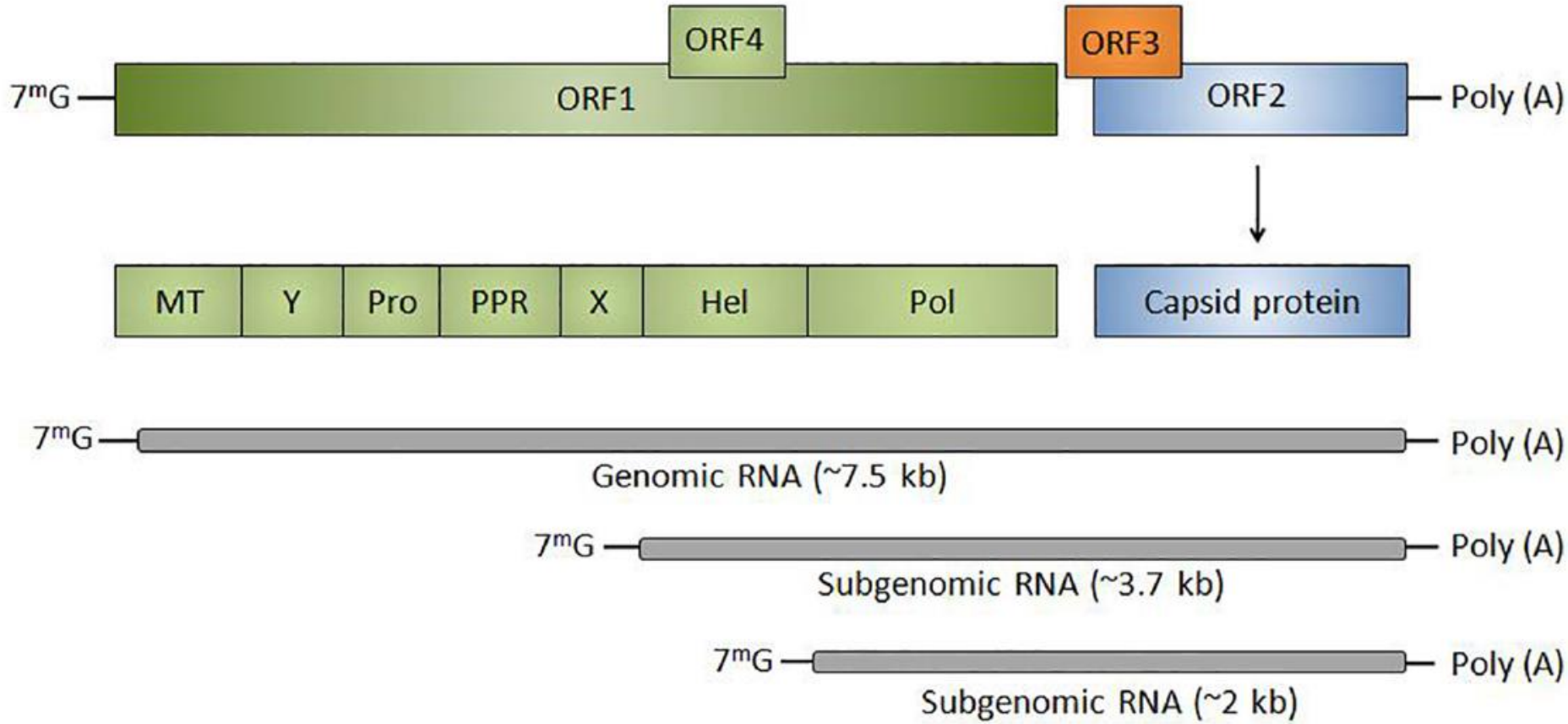

FIGURE 1 | Genomic organization of HEV: Classically divided into three ORFs (ORF1, ORF2, and ORF3) except genotype-1 having an additional ORF4. HEV RNA is transcribed into subgenomic RNA of ~3.7 and ~2 kb. The 5′ non-coding region is capped with 7-methylguanosine (7mG) and 3′ is polyadenylated [poly(A)]. ORF1 encodes for non-structural proteins including MT, regions of unknown function (Y, PPR, and X), Pro, Hel, and Pol or RdRp. ORF2 codes for capsid protein and ORF3 is a multifunctional protein that helps in the release of virions from the infected cell.

Sofosbuvir

- Sofosbuvir è un profarmaco nucleotidico, che può essere incorporato nell'HCV RNA dal RdRp NS5B che agisce come terminatore di catena. Il suo potenziale antivirale contro HEV è stato dimostrato utilizzando i repliconi HEV GT3 nelle cellule Huh7 e HepG2. I valori IC50 erano in un intervallo micromolare basso, da 1 a 2 ordini di grandezza meno potenti rispetto ai repliconi HCV. Sofosbuvir ha mostrato un effetto additivo insieme a RBV. L'effetto inibitorio è stato confermato dallo stesso gruppo in cellule staminali pluripotenti indotte derivate da epatociti (iPSC-HLC) per tutti e quattro i GT comuni e in coltura cellulare per GT1 e GT3 da altri gruppi. è stato descritto che sia i repliconi GT1 e GT3 che il virus GT3 a lunghezza intera non erano inibiti nelle cellule HEK293T, U-87MG e Huh7. Li et al. hanno riferito che un ceppo GT5 è stato moderatamente inibito a 200 M da un estere isopropilico di Sofosbuvir, PSI 7977.
- L'efficacia di Sofosbuvir in vivo è stata inconcludente. 3case ha dichiarato che Sofosbuvir non è riuscito a eliminare l'HEV quando somministrato insieme a RBV, mentre 3 ha riportato un trattamento di successo insieme a RBV. Uno studio multicentrico di fase II Sofosbuvir in monoterapia (400 mg una volta al giorno 24 settimane) in 9 pazienti precedentemente falliti RBV. Nessuno dei pazienti ha eliminato il virus, sebbene abbia ridotto i livelli di HEV RNA. potrebbe essere ulteriormente studiato in combinazione con RBV.
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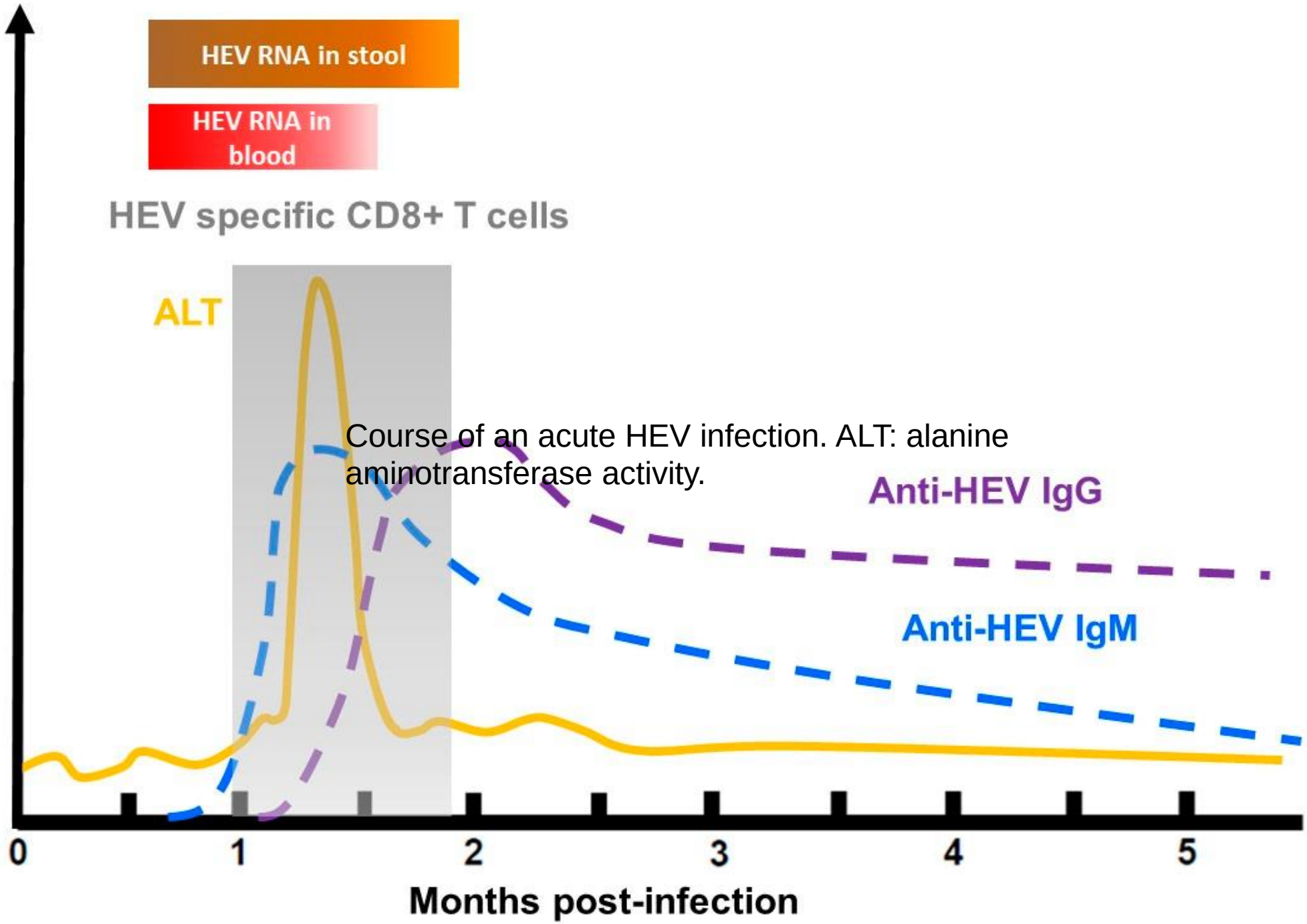
Public health control measures

- In December 2011, the first HEV recombinant subunit vaccine (Hecolin®) was registered in China for the prevention of HEV infection and HEV-related diseases in humans. So far, it has not been licensed and approved in any other country. Although randomized controlled trials have shown a high efficiency and very low number of SAEs following HEV vaccination, it is not recommended by WHO for routine use in under 16 years, pregnant women, people with chronic liver disease, people on organ transplant waiting lists, and travellers .
- The WHO position concerning routine vaccination programs should not preclude the use of the vaccine in specific situations such as outbreaks where the risk of hepatitis E or of its complications or mortality is particularly high.

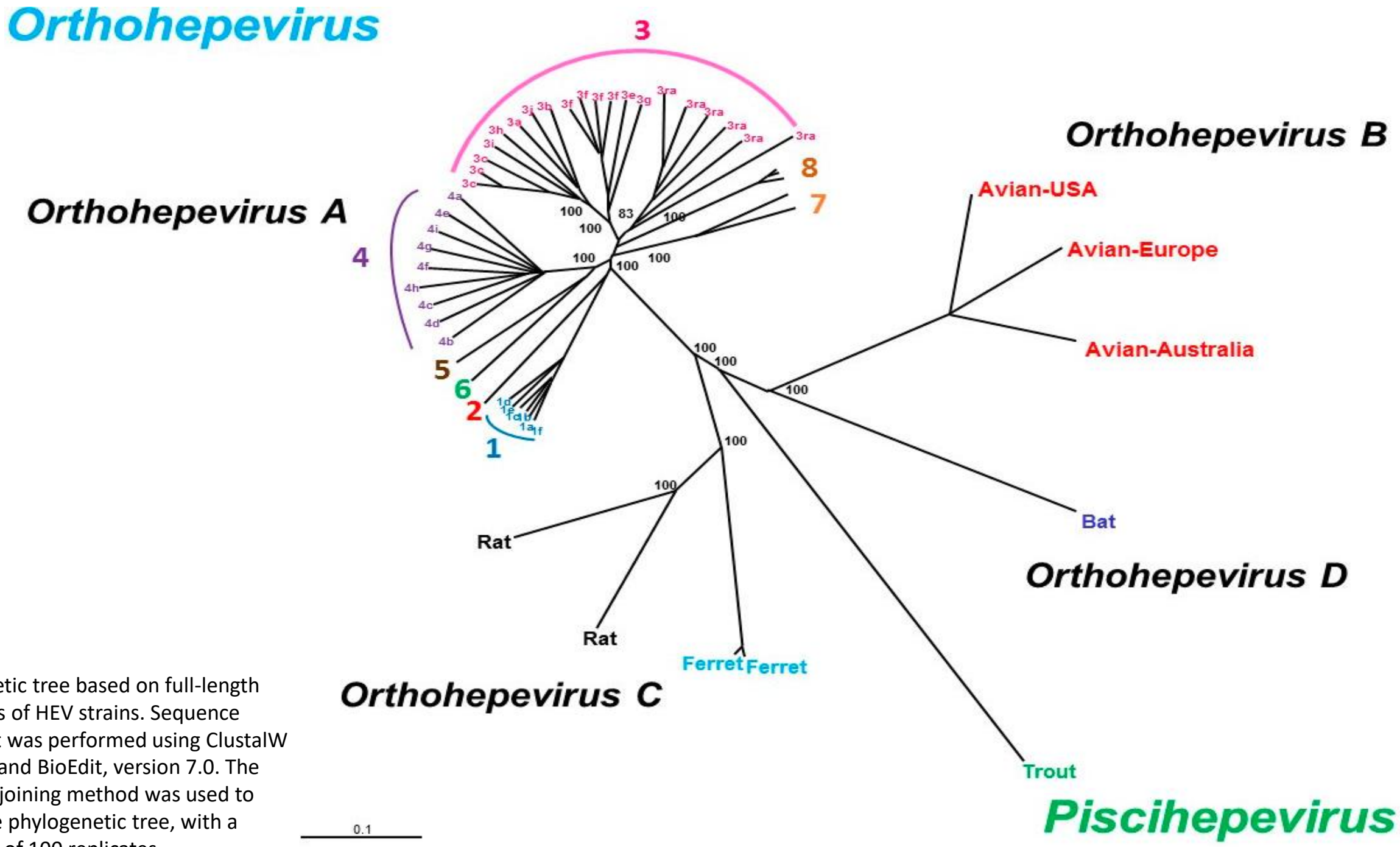
Infection control, personal protection and prevention of infection

- Consumption of raw or undercooked pig meat and shellfish should be avoided in all parts of the world. It is important to make sure that processed food containing pig meat is well cooked.
- When travelling to countries with poor sanitation, it is advisable to boil all drinking water, including water used for brushing teeth.
- Wearing working gloves and boots might reduce HEV infection in those workers with occupational exposure to HEV-infected animals.

- Transfusion or transplantation-related HEV transmission events have been described and the conduction of risk assessment is recommended for each product with special consideration of risk groups. Some assessments stated that transfusion-transmitted infection plays a minor role as source of infection in the risk group of solid-organ recipients and risk factors have been identified to be consumption of pork or game meat.



Course of an acute HEV infection. ALT: alanine aminotransferase activity.



Phylogenetic tree based on full-length sequences of HEV strains. Sequence alignment was performed using ClustalW (MEGA5) and BioEdit, version 7.0. The neighbor-joining method was used to create the phylogenetic tree, with a bootstrap of 100 replicates.

Year(s)	Country	Mode of Transmission	Reported
2013	Cameroon	Waterborne	33
2014–2015	Bangladesh	Waterborne	103
2014–2016	India	Waterborne	17
2016–2017	Chad	Waterborne	1293
2017–2018	Nigeria	Contamination of drinking water	1376
2014–2017	Bangladesh	Waterborne	661
2018	Central African Republic	Waterborne	149
2018	South Sudan	Waterborne	161
2019	Pakistan	Waterborne	300
2017–2020	Namibia	Waterborne	7247

[1] Numbers based on reported estimates.

Diagnostica

- Diagnosi AHE: IgM anti-HEV nel siero e/o HEV RNA nel siero o feci.
- Genoma HEV: PCR generica o RT-PCR.
- Metodi basati su Ab (Western blot o immunoassay enzimatici) CHE: IgG anti-HEV
- L'infettività dell'HEV è determinata nell'inoculazione sperimentale di animali o con tecniche di coltura cellulare.
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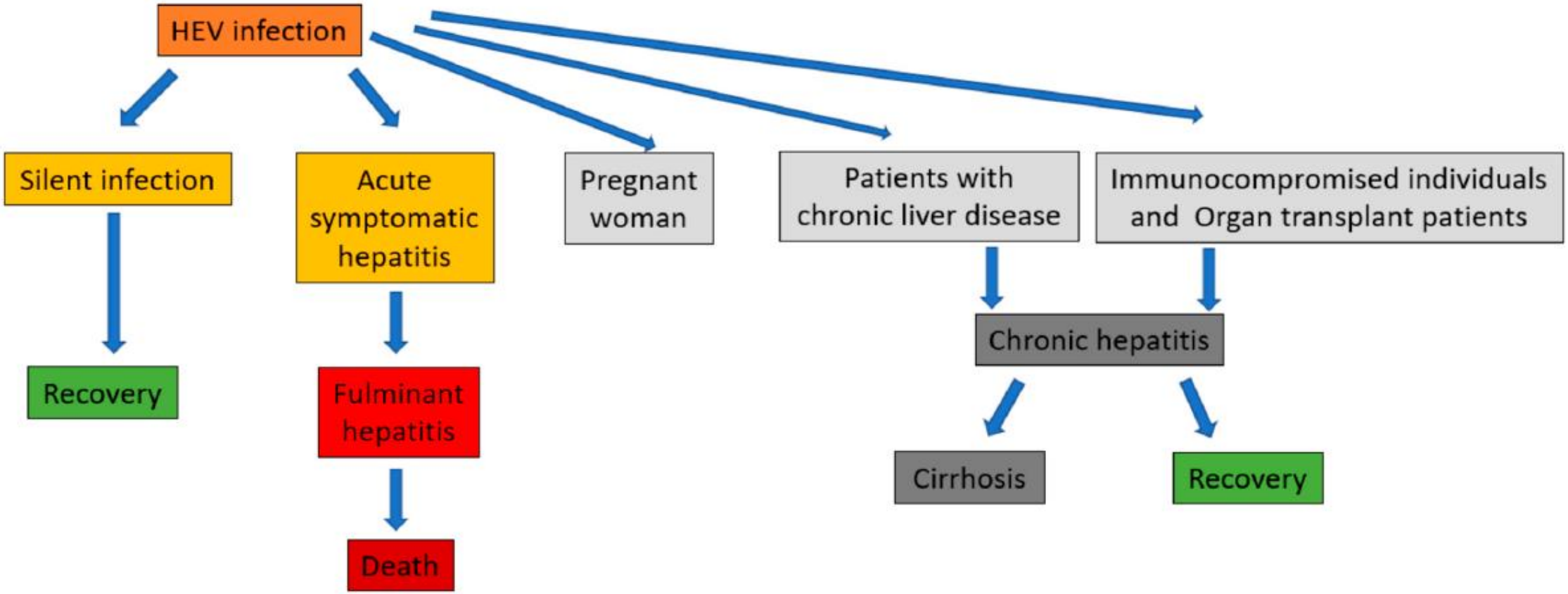
Different disease scenarios seen with HEV.



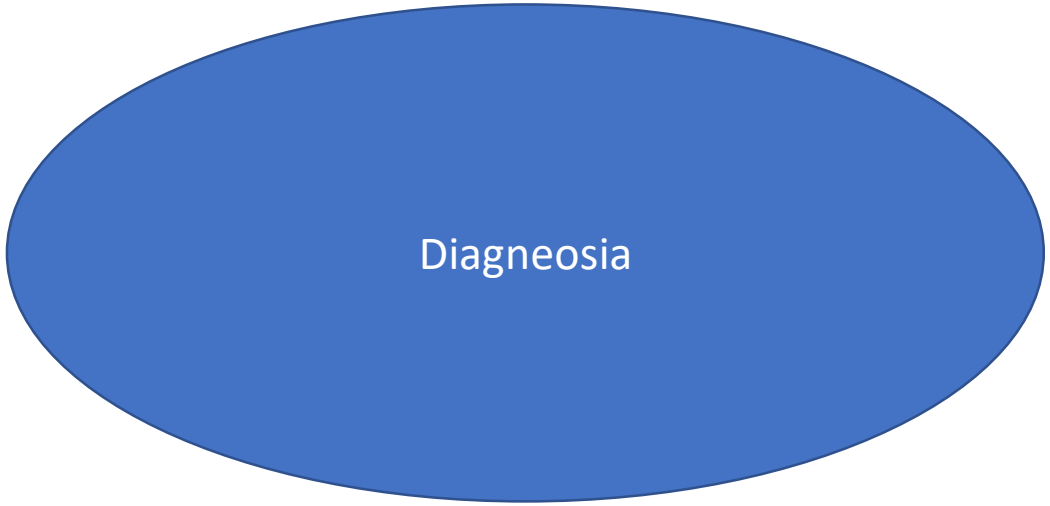

Diagneosia

Figure 8. Laboratory tests required to confirm acute case

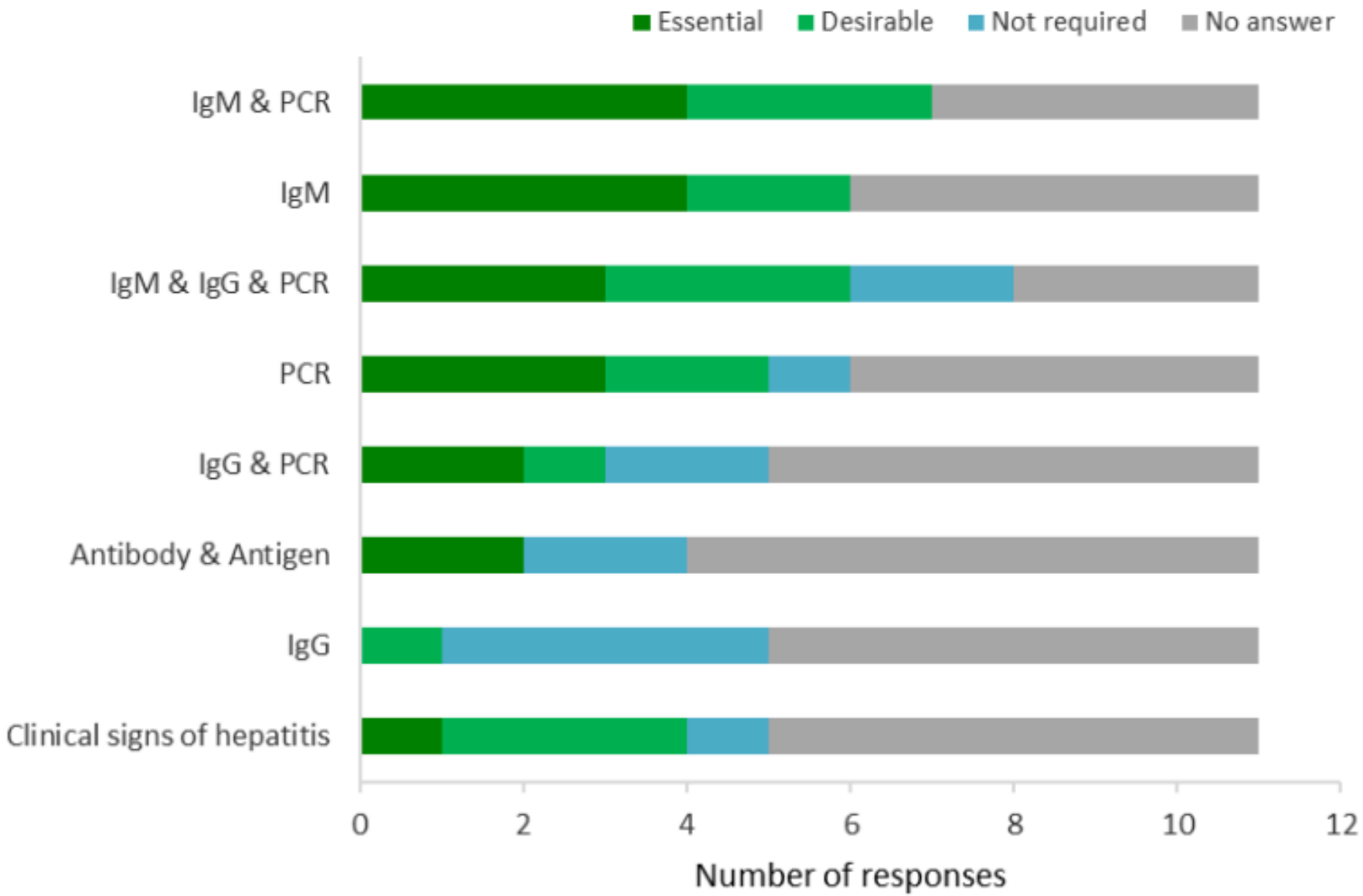
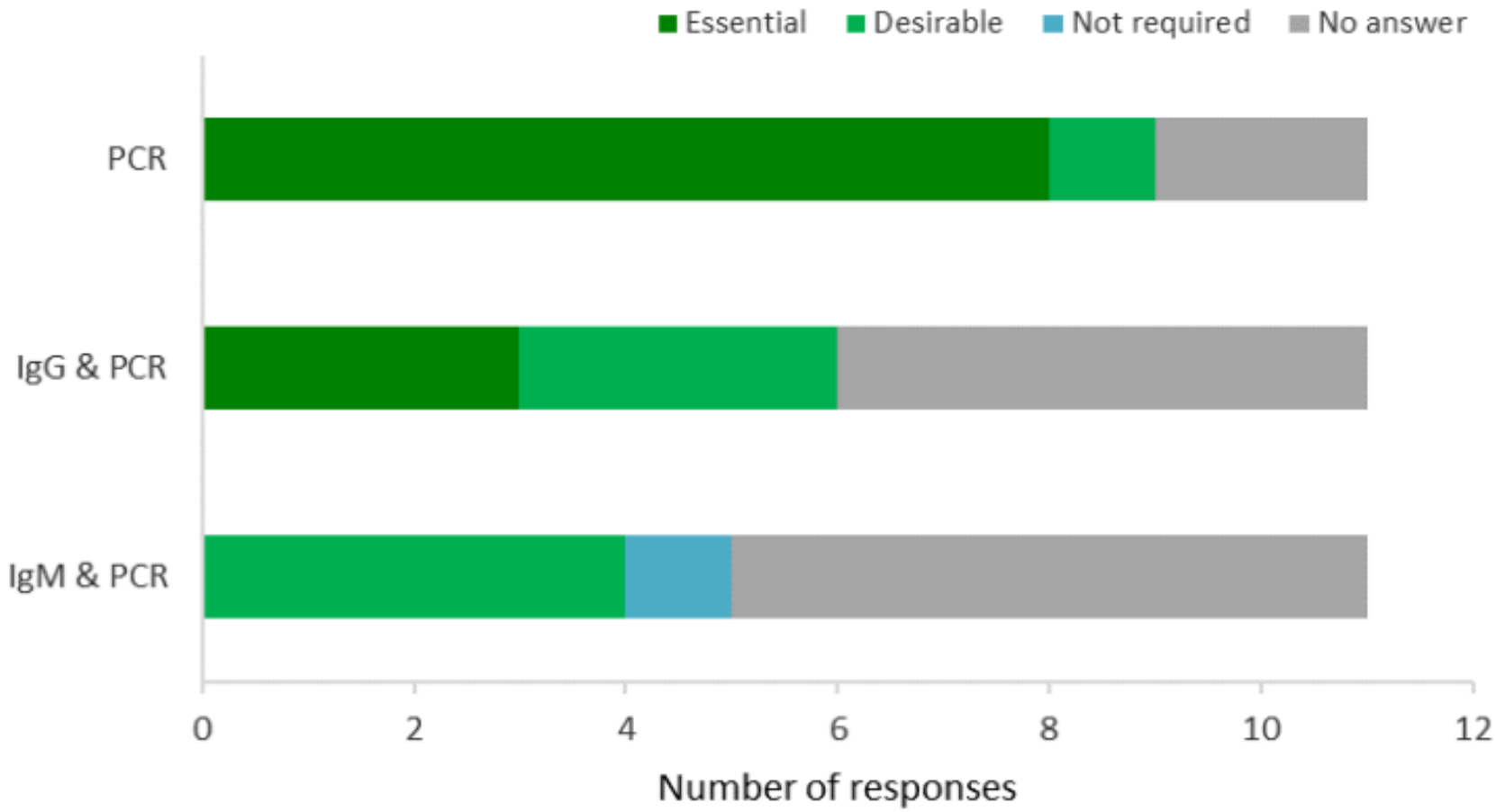


Figure 9. Laboratory tests required to confirm a chronic case



Chronic case minimum time period

There were eight responses to the question about minimum time period of infection to diagnose a chronic case:

- 3 months (four responses)
- 3–6 months (two responses); and
- 6 months (two responses).

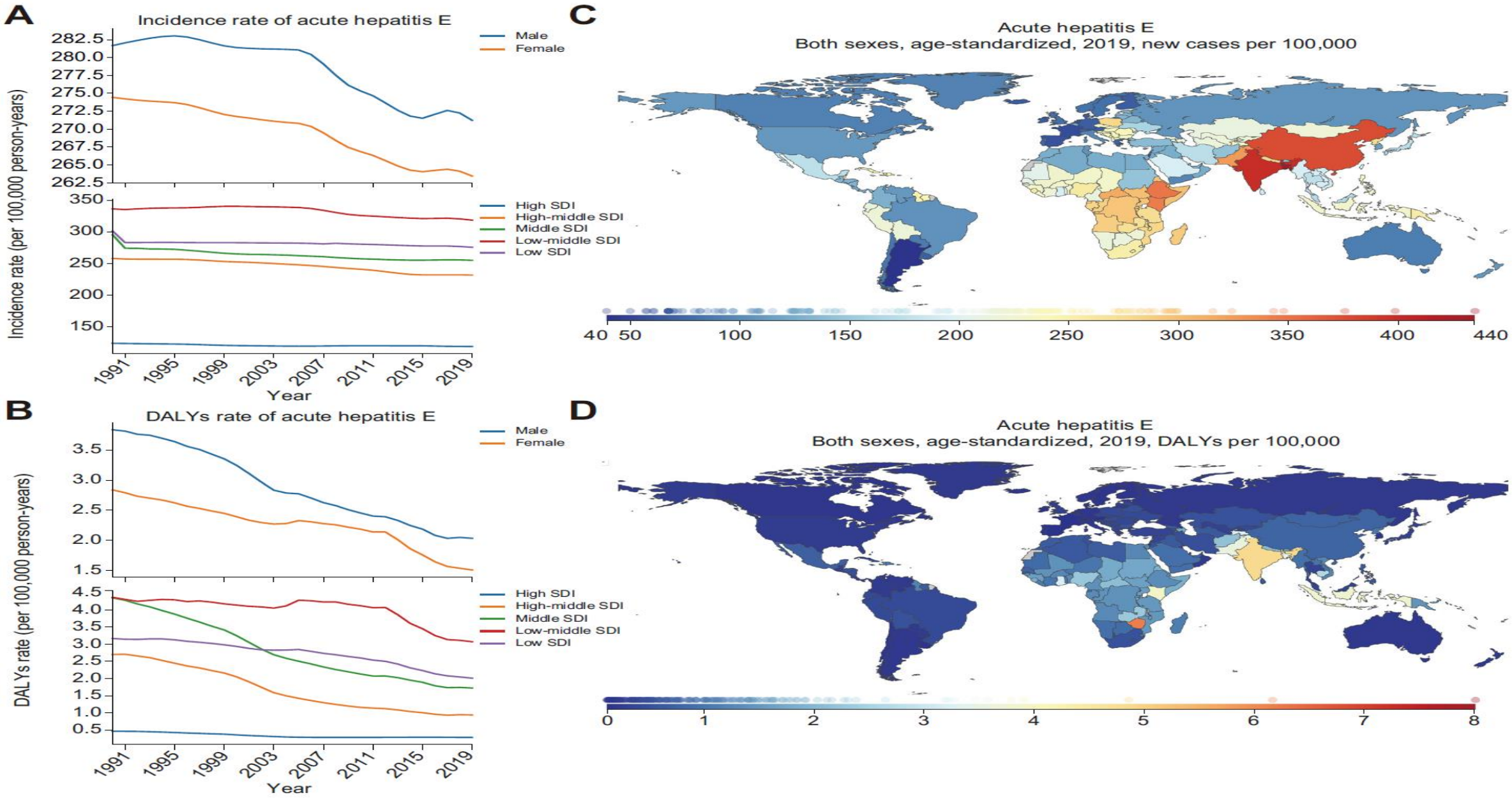


Fig. 5. Burden of AHE for 204 countries and territories. Age-standardized (A) incidence and (B) DALY rates of AHE per 100,000 population from 1990 through 2019 categorized according to country and territory, stratified by sex or SDI. Age-standardized (C) incidence and (D) DALY rates of AHE per 100,000 person-years by country and territory, in 2019. The maps in (C) and (D) were generated by the GBD 2019 tool. AHE, acute hepatitis E; DALY, disability-adjusted life year; GBD, Global Burden of Disease; SDI, socio-demographic index.

Legame clinico tra compromissione delle cellule linfoidi ed esito hev

- Pazienti dopo SOT
- Tacrolimus:f attore di rischio per il suo effetto immunosoppressivo più elevato rispetto alla ciclosporina A. Entrambi avevano un potere molto potente nel colpire le cellule T.
- oltre il 60% dei pazienti con HEV abbia sviluppato CHE in trapianto di cuore .
- solo l'acido micofenolico (MFA) era associato alla clearance HEV. MFA inibisce HEV.
- MFA mostra effetti antivirali diretti sulla replicazione dell'HEV; L'AMF deve essere presa in considerazione nei pazienti con HEV-SOT quando possibile, sebbene gli effetti specifici richiedano ulteriori indagini.
-

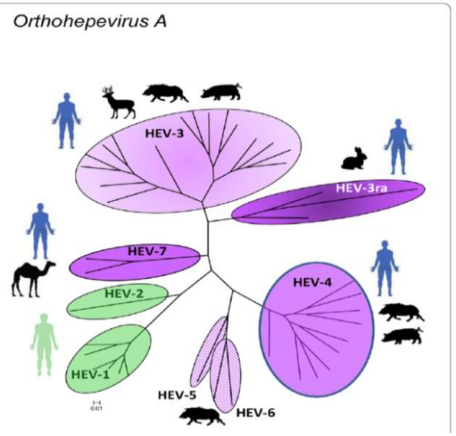
foods

MDPI

Article

Quantitative Methods for the Prioritization of Foods Implicated in the Transmission of Hepatititis E to Humans in Italy

Ornella Moro [1,2,*], Elisabetta Suffredini [1], Marco Isopi [2], Maria Elena Tosti [3], Pietro Schembri [4] and Gaia Scavia [1]



The food sampling survey indicated the highest proportion of HEV positive samples (i.e., HEV food prevalence αi) belongs to PL products (11%), followed by PNL (2.8%) and SH (0.48%).

No positive samples were found for tGV and RM categories (0)

Table 4. Mean viral load per serving (g.e./gr) $\mathbb{E}(C_1^i)$, meaning λ_i^{-1}, the average number of serving consumed in one year d_i^a and the prevalence of HEV positive food samples for each category α_i.

Category	$\mathbb{E}(C_1^i)$ (g.e/serving)	d_i^a (no.serving/year)	Prevalence α_i
PL	78,000	3	0.11
PNL	34,000	49	0.028
SH	87,000	26	0.0048
GV	0	132	0
RM	0	107	0

Donor-Derived Genotype 4 Hepatitis E Virus Infection, Hong Kong, China, 2018

Siddharth Sridhar, Vincent C.C. Cheng, Shuk-Ching Wong, Cyril C.Y. Yip, Shusheng Wu, Anthony W.I. Lo, Kit-Hang Leung, Winger W.N. Mak, Jianpiao Cai, Xin Li, Jasper F.W. Chan, Susanna K.P. Lau, Patrick C.Y. Woo, Wai-Ming Lai, Tze-Hoi Kwan, Timmy W.K. Au, Chung-Mau Lo, Sally C.Y. Wong, Kwok-Yung Yuen

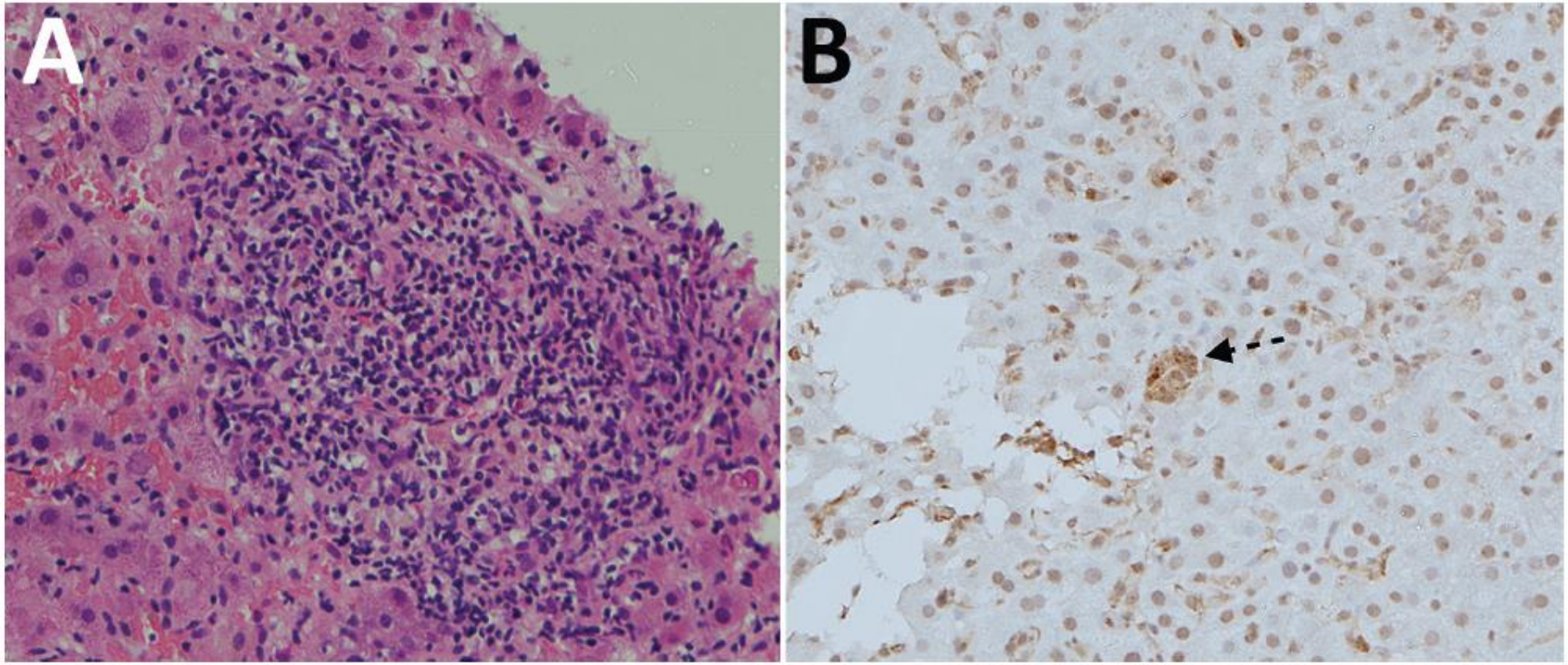
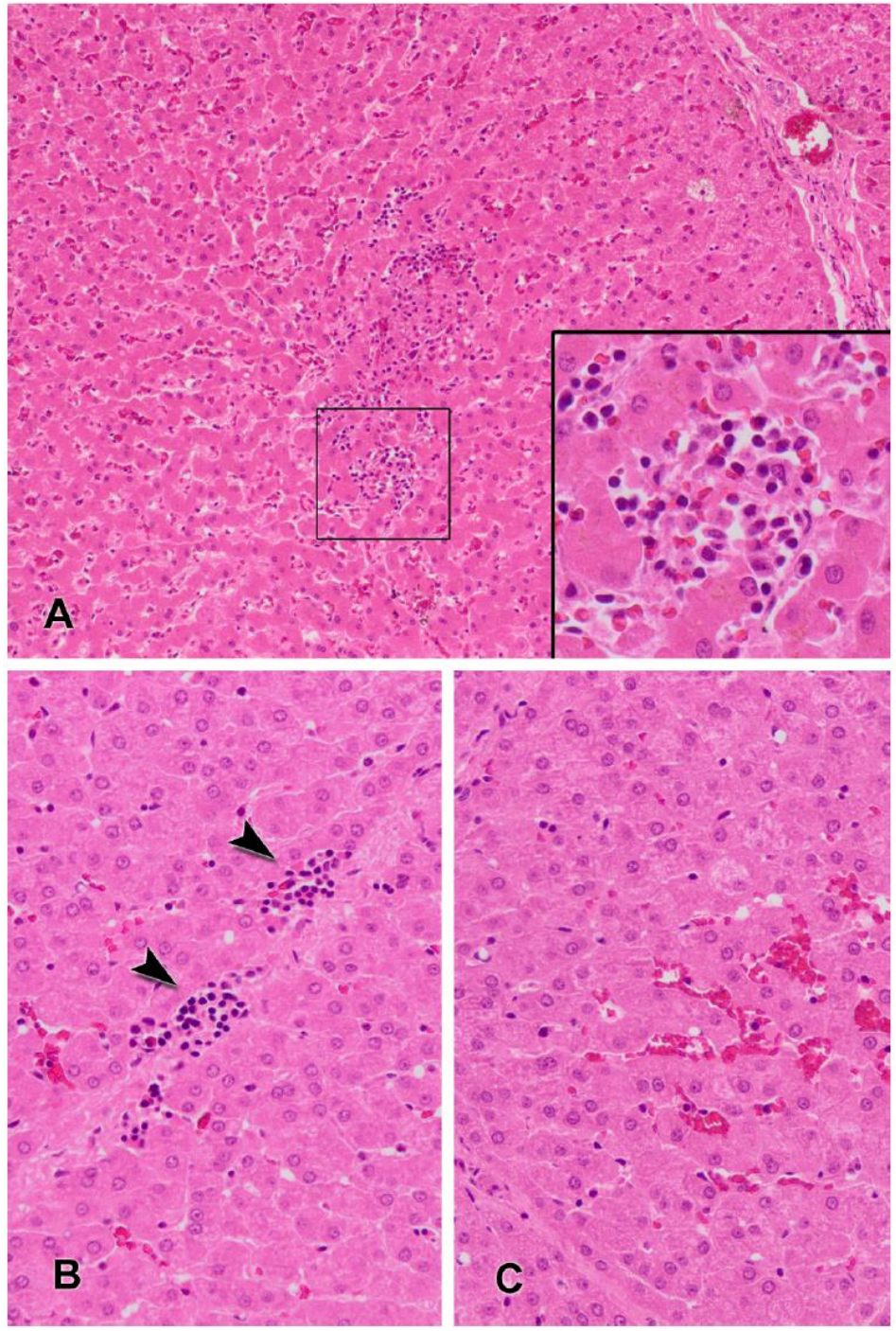


Figure 3. Histology of tissue from liver graft of hepatitis E virus case-patient 1, a 66-year-old man, on posttransplantation day 122. A) Hematoxylin and eosin staining showing moderate (grade 2) inflammation. B) Immunohistochemical staining (using hepatitis E virus monoclonal antibody); arrow indicates small groups of hepatocytes with positive cytoplasmic signals. Original magnification ×200.

Histopathologic lesions in wild boar naturally infected with HEV

- The livers of the 4 animals with naturally occurring HEV infection displayed only mild diffuse lymphoplasmacytic infiltrates (Fig 1A and 1B), with one of them also showing slight signs of hepatocyte necrosis. One of these animals also had a high degree of congestion in the hepatic sinusoids (Fig 1C).

- Fig 1A: liver with presence of mild intralobular lymphoplasmacytic aggregates in the hepatic parenchyma. Inset showing a lymphoplasmacytic aggregate at higher magnification (hematoxylin and eosin, HE).
- Fig 1B: liver showing slight perilobular infiltrations of lymphocytes (arrowheads) (HE).
- Fig 1C: hepatic lobule with moderate
- hyperemia of liver sinusoids (HE).



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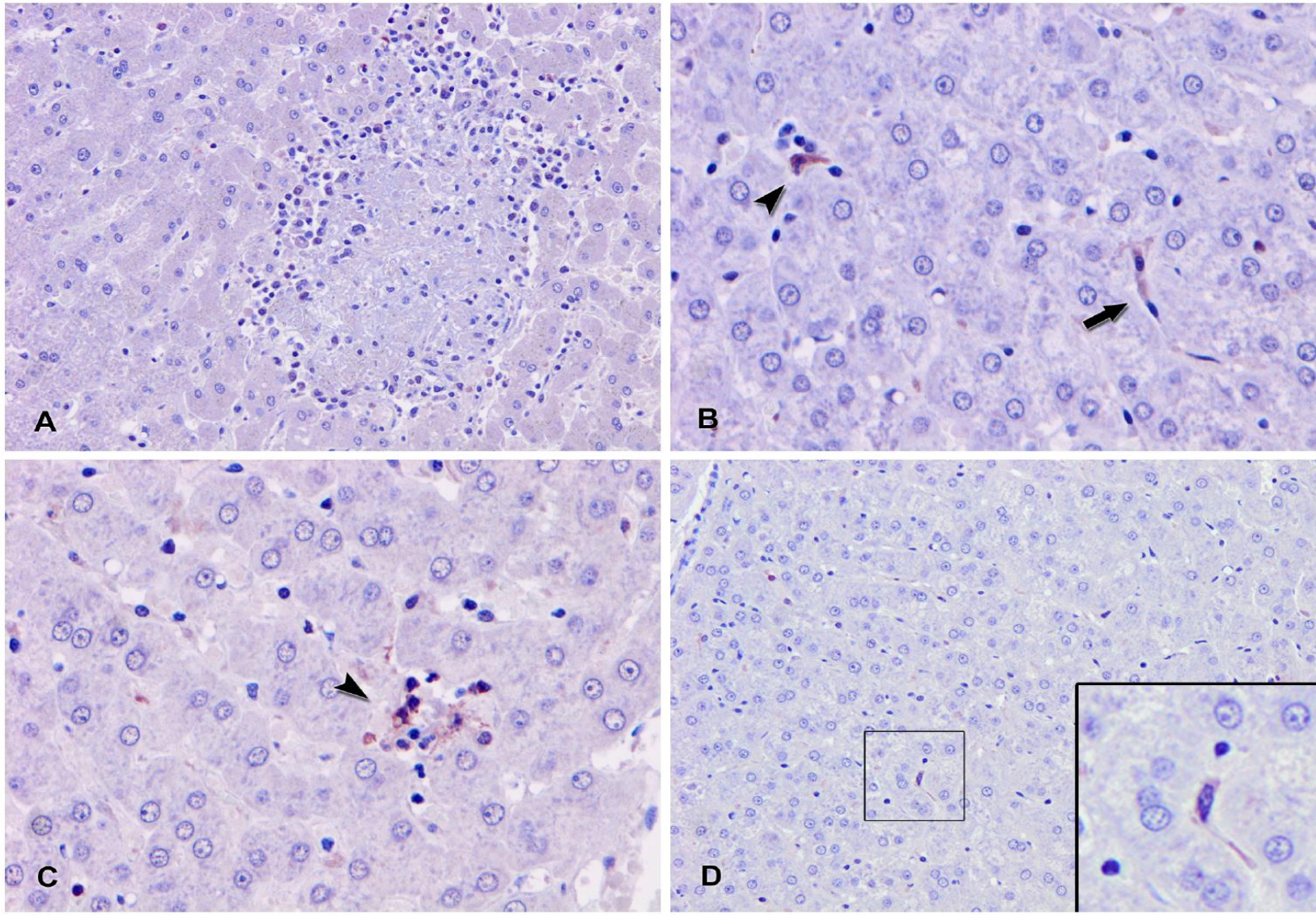
Representative photomicrographs of liver sections from 3 wild boar naturally infected with HEV

- Fig 2A: liver with absence of association of HEV antigen inside lymphoplasmacytic aggregates (Immunohistochemistry (IHC) with avidin-biotin-peroxidase complex (ABC) method counterstained with hematoxylin).
- Fig 2B: HEV immunolabeling in cytoplasm of Kupffer cells (arrowhead) and sinusoidal endothelial cells (arrow) (IHC ABC method).
- Fig 2C: HEV immunostaining of light lymphoplasmacytic infiltrate (arrowhead) (IHC ABC method).
- Fig 2D: liver showing slight labeling for HEV antigen in hepatic parenchyma, which is appreciable in Kupffer cells (inset) (IHC ABC method).



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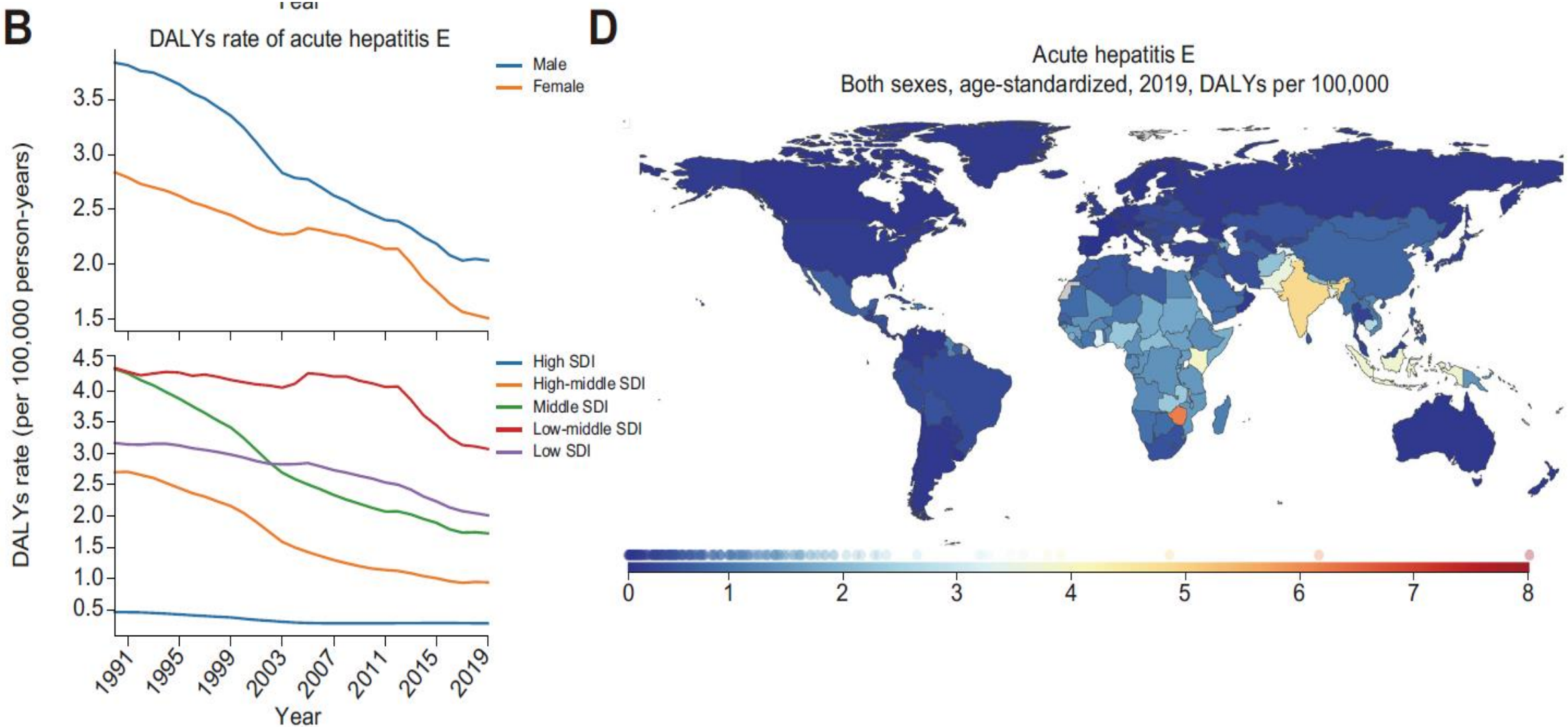

Fig. 5. Burden of AHE for 204 countries and territories. Age-standardized (A) incidence and (B) DALY rates of AHE per 100,000 population from 1990 through 2019 categorized according to country and territory, stratified by sex or SDI. Age-standardized (C) incidence and (D) DALY rates of AHE per 100,000 person-years by country and territory, in 2019. The maps in (C) and (D) were generated by the GBD 2019 tool. AHE, acute hepatitis E; DALY, disability-adjusted life year; GBD, Global Burden of Disease; SDI, socio-demographic index.