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Hepatitis C Virus Transmission in a Pediatric Oncology Ward: Analysis of an Outbreak and Review of the Literature

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SUMMARY: Hospital-related hepatitis C virus (HCV) infections continue to occur even after the introduction of blood donor screening. We report an outbreak of HCV in nine patients of a pediatric oncology ward in 1996/1997. Sequencing of the hypervariable genomic region 1 (HVR1) of the E2/NS1 region showed near identity between HCV isolates from these patients as evidence for infection with the same virus. Despite a detailed and careful investigation, the source of infection and the mode of virus transmission could not be established. Based on a review of the current literature about nosocomial HCV infection and HCV infection in children, hypotheses for possible means of transmission in this outbreak are discussed. (*Lab Invest 2001, 81:251–262*).

The discovery of the hepatitis C virus (HCV) in 1989 by Choo et al (1989) and the subsequent development of serological tests for HCV infection have shown that HCV is the principal cause of transfusion-associated and sporadic parenteral non-A, non-B hepatitis. Prevalence rates of infection are 0.1% to 1.5% in Europe, with a North-South gradient, 0.6% in the United States, 1% to 2% in China, Thailand, and Japan, and 0.2% to 20% in Africa (Botte and Janot, 1996).

It is estimated that chronic infection develops in about 85% of individuals (Rosen, 2000). In these patients, persistent or fluctuating alanine aminotransferase (ALT) levels indicating active liver disease are present in 60% to 70%. Cirrhosis develops in 10% to 20% over a period of 20 to 30 years, and hepatocellular carcinoma in 1% to 5%. In the United States, about 40% of chronic liver disease is HCV-related, resulting in 8000 to 10,000 deaths per year (Centers for Disease Control and Prevention, 1998). According to our present knowledge, HCV transmission occurs mainly through large or repeated direct percutaneous exposures to blood. Less frequent routes are sexual transmission (Alter et al, 1989), perinatal transmission (Ruiz-Moreno et al, 1999), and acquisition from mucous membrane exposure (Rosen, 1997; Sartori et al, 1993). However, in up to 40% of infected individuals, the route of transmission remains unknown (Alter, 1995). Injected drug use currently accounts for most HCV transmissions in the United States (Centers for Disease Control and Prevention, 1998).

Since the introduction of blood and organ donor screening by antibody testing in 1991, HCV has rarely been transmitted by transfusion of blood products, at least in developed countries (Centers for Disease Control and Prevention, 1998). However, hospital-related HCV transmissions continue to occur. The evaluation of this problem is difficult because of the frequently silent course of infection, the lack of prospective studies, and the scarce data about patient-to-patient transmission in settings other than hemodialysis units.

Herein, we report an outbreak of HCV involving nine patients in a pediatric oncology ward. We have analyzed all epidemiological aspects of this outbreak.

Case Presentation and Results

Presentation of the First Case

The first case (P1) identified in the described outbreak was a 2-year-old boy with intestinal malabsorption caused by idiopathic autoimmune enteropathy. He had been hospitalized since birth in a pediatric oncology ward for complete intravenous feeding. In July 1996, he developed acute hepatitis with jaundice, hepatomegaly, fatigue, and elevated ALT levels. HCV-RNA-PCR was positive in serum at this time, and anti-HCV-antibodies became positive 10 weeks later (Fig. 1). Serotyping and genotyping showed infection with HCV type 1b. The patient stayed in the oncology ward until February 1998 when he was moved to

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another ward. After a combined liver and small bowel transplantation in May 1999, he did not require intravenous feeding any longer and left the hospital. Since October 1997, he has been repeatedly HCV-RNA negative in serum.

The Outbreak

Repeated testing of the other patients of the oncology ward during the following months disclosed eight additional children (P2–P9) who became positive for HCV-RNA (Fig. 1). None of them showed clinical signs of acute hepatitis or significantly elevated ALT levels (Table 1). Testing for anti-HCV antibodies showed seroconversion to anti-HCV for seven children. Two patients (P5, P6) died from their oncologic disease before seroconversion. As in patient P1, serotyping and genotyping revealed HCV type 1b, the most common type in Germany.

Sequence comparisons of the hypervariable region HVR1 showed near identity between HCV isolates from patient P1 and 7 of the 8 newly infected patients compared with HCV isolates from 10 control patients with HCV type 1b infection from the same geographical area (Fig. 2). This finding was indisputable proof of a common source of infection. For patient P2, HCV-RNA-PCR repeatedly gave weak products not sufficient for sequence analysis. However, acute seroconversion to anti-HCV and infection with genotype 1b suggested that this patient was also infected with the same virus strain.

For six children, follow-up data for 24 months and longer were available: four developed chronic HCV infection with persistently positive HCV-RNA-PCR in serum and slightly elevated ALT levels. In patient P9, liver biopsy was performed in June 1999 and showed chronic hepatitis with fibrosis (portal inflammation grade 1, lobular inflammation grade 1, and fibrosis stage 2 according to the histological evaluation proposed by Yano et al, 1996). Two patients (P2, P7) probably cleared HCV infection and have remained HCV-RNA-PCR negative since July 1997. ALT levels were in the normal range.

Additional Single Cases

Before the first admission to the oncology ward, all patients had routinely been screened for the presence of anti-HCV antibodies. In 1996 and 1997, only two patients (P10, P11) were anti-HCV-positive already at this time. However, they were repeatedly negative by HCV-RNA-PCR, and genotyping or sequence analysis could not be performed. Serotyping gave no unequivocal results for patient P10 and showed HCV type 3 for patient P11.

Search for Possible Transmission Routes

When the first case of hepatitis C (P1) was recognized, screening of the parents by anti-HCV antibody testing and HCV-RNA-PCR gave negative results, and the parents and a later-born sister remained negative at a follow-up investigation in 2000. Stool and vomit of patient P1 were investigated and were found to be negative by HCV-RNA-PCR. Thus, virus transmission by possibly still unrecognized and unusual routes to

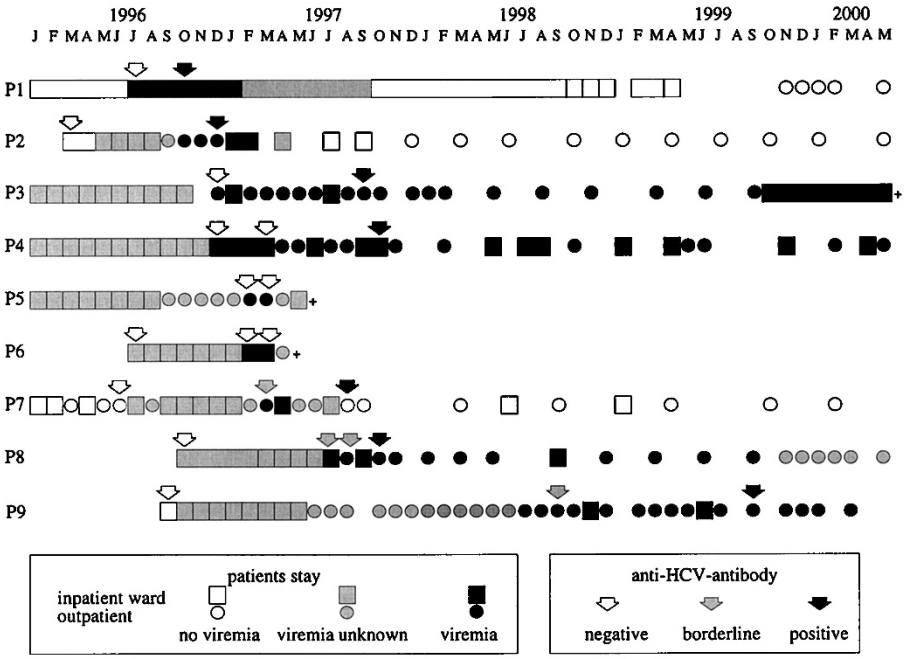

Figure 1.

Treatment periods at the pediatric oncology department during the outbreak of hepatitis C virus (HCV) genotype 1b. Patients are designated P1 to P9. Treatment periods are shown by *boxes* (inpatient) and *circles* (outpatient). White and black symbols represent periods during which serum samples were HCV-PCR negative and positive, respectively. Gray symbols denote periods during which no serum was available to determine viremia status. *Arrows* show results of anti-HCV antibody testing. *Crosses* (+) show dates of death.

Table 1. Patients Newly Infected with Hepatitis C Virus (HCV) Genotype 1b

Patient	Age[a] (yr)	Sex	Diagnosis	Chemotherapy[a]	Symptoms of acute hepatitis	Follow up for ≥ 48 mo
P1	2	M	Autoimmune enteropathy	Yes	Hepatomegaly, jaundice, fatigue, ALT elevation	HCV-RNA negative ALT normal
P2	1	F	Nephroblastoma	Yes	None[b]	HCV-RNA negative ALT normal
P3	4	M	Neuroblastoma	Yes	None[b]	HCV-RNA positive ALT slightly elevated[c]
P4	3	F	Medulloblastoma	Yes	None[b]	HCV-RNA positive ALT slightly elevated[c] Died 7/00[d]
P5	2	M	Medulloblastoma	Yes	None[b]	Died 6/97[d]
P6	0.5	M	Leukemia	Yes	None[b]	Died 4/97[d]
P7	3	F	Nephroblastoma	Yes	None[b]	HCV-RNA negative ALT normal
P8	1	M	Lymphangioma	Yes	None[b]	HCV-RNA positive ALT slightly elevated
P9	10	M	Malignant lymphoma	Yes	None[b]	HCV-RNA positive ALT slightly elevated[e]

ALT, alanine aminotransferase.
[a] When contracting HCV in 1996/97.
[b] ALT levels slightly elevated but not higher than expected with chemotherapy.
[c] With continuing chemotherapy.
[d] Died from malignant disease.
[e] Liver histology (June 99) showed chronic hepatitis with fibrosis: portal inflammation grade 1, lobular inflammation grade 1, and fibrosis stage 2 according to the histological evaluation proposed by Yano et al (1996).

the other immunocompromised patients of this ward could be excluded. All hygienic precautions known to prevent HCV transmission were reinforced, eg, use and immediate change of disposable gloves for every percutaneous procedure, special care and coverage of the catheter entry site, and careful cleaning and disinfection of surfaces and nondisposable items.

The inpatient ward accommodates eight children in four two-bed rooms. Study of admission data and times of stay in the ward showed that all nine patients involved in this outbreak were being treated at overlapping periods. All nine children had a permanent central venous catheter for injections, infusions, and blood sampling. Multidose vials of saline were routinely used to flush catheters, but were not stored in the patients' rooms to prevent contamination with a previously used syringe. Staff members steadfastly denied any breaching of standard hygienic precautions.

Eight of the nine children had received blood products. Patient P1 had received varicella immunoglobulin about 4 months before infection. Retrospective testing of this immunoglobulin batch by HCV-RNA-PCR gave negative results. Three batches of blood products (immunoglobulin, antithrombin 3, albumin) were shared between patients but can be excluded as a common source of infection because they were administered after identification of the first HCV infection. All staff members were negative for anti-HCV. Except for the mother of P3, all parents and most of the siblings of patients P1 to P9 were tested and were found to be negative for anti-HCV.

In 1996 and 1997, 126 children were being treated in the pediatric oncology ward. Before admission, all patients were routinely tested for the presence of anti-HCV antibodies, and two patients (P10, P11) were found to be anti-HCV positive. Patient P10 was HCV-RNA negative and had possibly cleared the virus but cannot definitely be excluded as the source of infection. Patient P11 was infected with HCV serotype 3 and was therefore not involved in this outbreak. Of the remaining 115 patients who were being treated in the inpatient ward in 1996 and 1997, none showed symptoms of acute hepatitis. Serum samples were available for follow-up anti-HCV screening in 1998 to 2000 for 59 of these patients. No additional cases of HCV infection were detected.

Some limitations of this investigation deserve mention: (a) not all family members were investigated, (b) multidose vials, needles, or syringes were not available for testing, (c) not all patients of the oncology ward had been tested for anti-HCV after treatment. Newly infected children might have been missed.

Because we failed to identify the source and mode of transmission of HCV in this outbreak, a search of the actual literature about nosocomial HCV transmis-

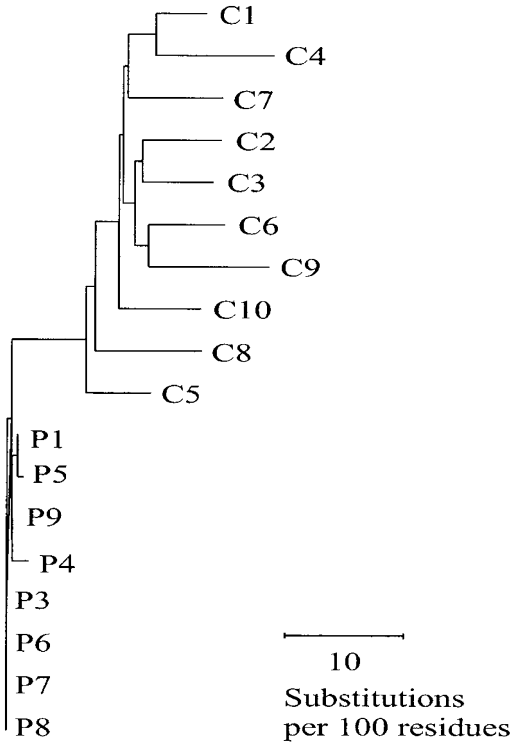

Figure 2.
Phylogram of HCV hypervariable region 1 of the hepatitis C virus genome (HVR1) sequences, created with DISTANCES and GROWTREE of the Genetics Computer Group (GCG) program package using the Kimura-2-parameter matrix and Neighbor joining. HCV isolates from patients involved in an outbreak of HCV (genotype 1b) are designated P1 and P3 to P9. HCV isolates from control patients infected with HCV genotype 1b are designated C1 to C10.

sion, ie, HCV transmission in the medical environment, and about HCV infection in children was performed.

Nosocomial Transmission of Hepatitis C Virus

HCV Transmission by Transfusion

The introduction of blood donor screening for antibodies against HCV in 1991 has greatly reduced the risk of posttransfusion hepatitis to an estimated residual risk of about 1 in 100,000 (Schreiber et al, 1996). More sensitive nucleic acid amplification tests can further reduce, but not completely eliminate, this risk as is demonstrated by a recent HCV transmission by a blood donation negative by HCV-RNA-PCR (Schuttler et al, 2000). Nevertheless, in industrialized countries blood transfusion can today widely be ruled out as a source of HCV infection.

Nontransfusional Transmission in Hemodialysis Settings

HCV infection of hemodialysis patients is the best known example of nosocomial HCV transmission other than by transfusion. These patients show a significantly higher prevalence of HCV infection than the general population. Prevalence rates vary widely between countries and among units within the same country, ranging between 8% and 39% in North America, 1% and 54% in Europe, 17% and 51% in Asia, and 1% and 10% in Australia (Sanchez-Tapias, 1999). The introduction of blood donor screening and erythropoietin treatment of anemia in dialysis patients has led to a fall in both the incidence and prevalence rates of HCV infection (Simon et al, 1994). Several observations have provided evidence for nontransfusional transmission: (a) length of time on hemodialysis was shown to be an independent risk factor for infection (Kapoor et al, 1993; Pujol et al, 1996; Salama et al, 2000); (b) infection of patients with no other parenteral risk factors than hemodialysis has been observed (Cendoroglo et al, 1995; DeLamballiere et al, 1996; Jadoul et al, 1993; Kapoor et al, 1993; Katsoulidou et al, 1999; Martin et al, 1993; Medin et al, 1993; Okuda et al, 1995; Pujol et al, 1996; Seme et al, 1997; Simon et al, 1994); (c) the risk of acquiring HCV infection was directly related to the prevalence of HCV antibody-positive patients being treated at some units (Kobayashi et al, 1998); (d) newly infected patients have often shown overlapping hemodialysis history (dialysis at the same time, during the same shift, in the same area) with HCV-positive patients (Allander et al, 1994; Irish et al, 1999; Izopet et al, 1999; Jadoul et al, 1993; Katsoulidou et al, 1999; McLaughlin et al, 1997; Norder et al, 1998; Okuda et al, 1995), and (e) there has been a lower prevalence (Chan et al, 1991; Pascual et al, 1993) and incidence of HCV infection in peritoneal dialysis (Cendoroglo et al, 1995; Medin et al, 1993).

Recently, a variety of studies have provided, by sequence analysis, reliable molecular evidence for patient-to-patient transmission (Abacioglu et al, 2000; Allander et al, 1994; DeLamballiere et al, 1996; Grethe et al, 2000; Hosokawa et al, 2000; Irish et al, 1999; Izopet et al, 1999; Katsoulidou et al, 1999; LePogam et al, 1998; McLaughlin et al, 1997; Norder et al, 1998), but the exact mode of transmission could not be established. In most cases, breaching or neglecting of hygienic precautions was suspected. For instance, 85% of Dutch centers allowed nurses to operate dialysis machines with gloves that were possibly contaminated with blood (Schneeberger et al, 1998). Gloves were the suggested mode of transmission in outbreaks in Greece (Katsoulidou et al, 1999) and Japan (Okuda et al, 1995). These authors speculated that the estimated 1000 touchings of the large needle skin hole for each patient per year provided entrance for the virus. In a Saudi Arabian dialysis unit with a 50% HCV prevalence, HCV-RNA was detected by PCR in handwashings from dialysis staff: in 24% of 80 samples from nurses dialyzing HCV-positive patients, in 8% of 100 samples from nurses dialyzing HCV-negative patients, and in 3% of 60 samples collected from personnel before entering the dialysis unit (Alfurayh et al, 2000). This emphasizes that the hands of dialysis personnel are a potential vehicle of HCV transmission.

Some authors reported that reinforcing strict hygiene, eg, the use of gloves whenever patients or hemodialysis equipment is touched, care in the use and disposal of needles and other sharp instruments,

no sharing of supplies, instruments, and medications among any patients, and the education of staff and patients, could significantly reduce or abolish new cases of infection (Izopet et al, 1999; Jadoul, 1995; LePogam et al, 1998; McLaughlin et al, 1997; Okuda et al, 1995, 1998; Seme et al, 1997). The reverse also holds true. For example, the constraints of the Gulf War led to an increase of anti-HCV prevalence in Kuwait from 30% to 71%, possibly by an increased spread of HCV as a result of the disruption of the normal hemodialysis environment (Kapoor et al, 1993).

Currently, there is no consensus regarding machine-sharing between HCV noninfected and infected patients. Although transmission by the same equipment has been suggested (LePogam et al, 1998; Simon et al, 1994), transmission has clearly been documented between patients who never shared equipment (McLaughlin et al, 1997). Two prospective studies showed that strict hygiene without separating machines was sufficient to prevent or significantly reduce new infections (Aucella et al, 2000; Gilli et al, 1995).

Finally, Zeuzem et al (1996) found evidence against patient-to-patient transmission by sequence comparisons between HCV isolates from 14 dialysis patients and 56 unrelated patients with chronic hepatitis C from the same geographical area. Therefore, other known (eg, via transfusion of blood products) and unknown transmission routes for HCV cannot be excluded.

Nontransfusional Transmission in Other Hospital Settings

There are only a few reports about patient-to-patient transmission outside hemodialysis wards (Table 2). Allander et al (1995) reported patient-to-patient transmission in a Swedish hematology ward; sequence analysis showed five clusters of closely related virus strains in 30 patients. The authors could not establish the mode of transmission and suggested unintentional mistakes in percutaneous procedures or other still unrecognized routes between immunocompromised patients. Rieske et al (1998) reported HCV infection in 21 children who underwent immunosuppressive therapy mainly for malignant disease in Germany. Analysis of all available data led to the conclusion that the infections were of nosocomial origin, although this was not proven by sequence comparison and the mode of transmission could not be identified. Widell et al (1999) described two HCV outbreaks in one pediatric oncology ward, each involving 10 patients in 1991 and 1993, respectively. Sequence analysis confirmed patient-to-patient transmission, and contamination of multidose vials was the suspected mode of transmission.

Schvarcz et al (1997) reported nosocomial HCV transmission from a subject with previously unrecognised chronic infection to two healthy subjects during a research project in a Swedish hospital. Faulty nursing and nonadherence to universal precautions were identified as probable causes of transmission. There is one report about HCV transmission during in-vitro fertilization (IVF) to two young women who had follicular puncture immediately after an HCV-infected woman (Lesourd et al, 2000). Sequence analysis proved nosocomial transmission. However, the mode of transmission could not be identified and the authors speculated that the contamination occurred outside the direct practice of IVF and possibly through procedures practiced by ancillary staff.

HCV transmission from seropositive patients to other patients at the time of colonoscopy has been reported twice (Bronowicki et al, 1997; LePogam et al, 1999). The main hypothetical routes of transmission may have been the use of the same colonoscope without adequate disinfection or the use of a multidose anesthetic vial or the same syringe. Obvious breaching of standard precautions when reusing syringes led to the transmission of HCV and human immunodeficiency virus during ozone therapy in Germany (Robert Koch Institut, 1997).

Transmission from Health Care Worker to Patient

Currently, there are only five reported HCV transmissions from health care workers to patients. A cardiac surgeon in Spain transmitted the virus to at least 5 patients (out of 222 patients investigated) between 1988 and 1994 (Esteban et al, 1996). Interestingly, transmission occurred during valve replacement surgery only when this surgeon performed the procedure himself, but not when he assisted other surgeons. The authors concluded that transmission was associated with percutaneous injury, most of which occurred during wire closure of the sternum. Two further cases of HCV transmission, one from a cardiac surgeon to 1 patient (Anonymous, 1995) and one from a gynecologist to 1 patient (Brown, 1999), have been reported without details. In Spain, an anesthesiologist infected 171 patients during the period 1994 to 1998 (Bosch, 2000). The issue is still sub judice, but it seems that this person was an addict and had given part of opioid analgesics to himself before giving the rest to his patients using the same syringe. A German anesthesiology assistant, who contracted infection from a patient, transmitted the virus to 6 patients during the incubation period before falling ill from acute hepatitis C (Ross et al, 2000). The virus was probably spread via a wound on the third finger of the assistant's right hand because he did not wear gloves when performing percutaneous procedures.

Transmission from Patient to Health Care Worker

The risk associated with occupational exposure to HCV is much lower than for hepatitis B virus. This has been suggested by studies that showed no higher prevalence in health care workers (0.28–2.0%) compared to the general population (Campello et al, 1992; Moens et al, 2000; Polish et al, 1993; Struve et al, 1994; Thomas et al, 1993; Zuckerman et al, 1994). However, the risk is not negligible, and large incidence studies have found that health care workers were two to three times more likely to develop non-A, non-B hepatitis than were controls (Lanphear et al, 1994;

Stroffolini et al, 1996). Hospitalized patients may serve as a reservoir for transmission, and the prevalence of anti-HCV seropositivity among patients can be as high as 18% in U.S. inner-city emergency departments (Kelen et al, 1992) or even higher in special hospital settings, eg, hemodialysis wards (Wreghitt, 1999).

Therefore, certain groups of health care workers may especially be at risk. Klein et al (1991) found significantly more dentists (1.7%) than blood donors (0.14%) to be seropositive for HCV, the highest rate (9%) was seen among oral surgeons. Higher prevalence rates have been reported for surgeons (4.3%) (DeMercato et al, 1996) and for health care workers involved with liver transplantation (5.3%) (Goetz et al, 1995) or working in internal medicine, pathology, or intensive care units (up to 7.1%) (Mihaly et al, 1996).

The most important risk factor appears to be unintentional needlestick injury, and transmission of HCV by this route has been confirmed using sequence comparisons of HCV isolates (Mizuno et al, 1997; Suzuki et al, 1994). The average incidence of anti-HCV seroconversion after needlestick injuries or exposure to sharps from an anti-HCV-positive source is 2% (range, 0–9.5%) (Table 3). Puro et al (1995) reported that transmission occurred only from hollow-bore needles but not from injuries with other sharp objects. Transmission is likely related to viral load, and Mitsui et al (1992) found a 10% HCV incidence after needlestick accidents involving an HCV-RNA-positive source.

Finally, HCV infection following conjunctival blood exposure was reported in two health care workers and in one employee of a state penitentiary (Ippolito et al, 1998; Rosen, 1997; Sartori et al, 1993). In one case, human immunodeficiency virus was transmitted simultaneously.

Other Possible Routes of Transmission

HCV-RNA has been found by PCR in many body fluids, including sweat, urine, and saliva (Liou et al, 1992; Nakano et al, 1992). The concentration of HCV in saliva seems to be several orders of magnitude lower than the concentration in blood (Taliani et al, 1997). Only a single case of HCV infection has been attributed to salivary transmission in the recipient of a human bite (Dusheiko et al, 1990). In this case, however, there was only circumstantial evidence for virus transmission, and the suspected source of infection was not tested for hepatitis C markers. In a recent systematic review, Ackerman et al (2000) found a low rate of intrafamilial transmission of HCV in family members of multitransfused, hemodialysis and hemophiliac patients with hepatitis C. Therefore, body fluids might be vehicles for virus transmission, but the efficiency of transmission seems to be quite low.

Hepatitis C in Children

Epidemiology

The prevalence of HCV infection in children is relatively low, with anti-HCV prevalence rates of 0.1% to 0.4% in the Western world, compared with a rate in the general population of 0.5% to 2% (Ruiz-Moreno et al, 1999). Before the availability of

Table 2. Previously Reported Cases of Nosocomial HCV Transmission (Outside Hemodialysis)

Reference	Transmission, setting	Suspected transmission route	Recommendations for prevention
Allander et al, 1995	Patient to patient, oncology	Percutaneous procedures, multidose vials	Adherence to infection control measures Restricted use of multidose vials
Rieske et al, 1998	Patient to patient, pediatric oncology	Unclear	Adherence to infection control measures Isolation of patients
Widell et al, 1999	Patient to patient, pediatric oncology	Multidose vials	Restricted use of multidose vials
Schvarcz et al, 1997	Subject to subject, research project	Flushing of IV catheters, contaminated gloves	Use of syringes only once Change of gloves between subjects
Lesourd et al, 2000	Patient to patient, in vitro fertilization	Procedures performed by ancillary staff (not clarified)	Exclusion of HCV-RNA-positive patients from in vitro fertilization
Bronowicki et al, 1997; LePogam et al, 1999	Patient to patient, colonoscopy	Endoscope, multidose vials	Adherence to disinfection protocol for endoscopes Restricted use of multidose vials
Robert Koch Institut, 1997	Patient to patient, ozone therapy	Reuse of syringe to inject ozone into patient's blood	Adherence to instructions for use and to infection control measures
Esteban et al, 1996	Surgeon to patient, cardiac surgery	Percutaneous injury (wire closure of sternum)	Prevention of injury-prone procedures in infectious health care workers
Bosch, 2000	Physician to patient, anesthesiology	Addicted physician injected part of opioid analgesics to himself and rest to patients	No sharing of syringes
Ross et al, 2000	Patient to nurse to patient, anesthesiology	Percutaneous procedures performed without gloves by nurse with open wound on finger	Adherence to infection control measures (use of gloves)

Table 3. Risk of HCV Infection in Anti-HCV-Negative Health Care Workers after Occupational Exposures to an Anti-HCV-Positive Source[a]

		Accidents		HCV infection	
Reference	Follow up (mo)	n	Type	n	Rate (%)
Kiyosawa et al, 1991	≥6	110	Needlestick	3	2.7
Hernandez et al, 1992	≥6[b]	81	Needlestick	0	0
Mitsui et al, 1992	≥6	68	Needlestick, HCV-RNA positive source	7	10.3
		8	Needlestick, HCV-RNA negative source	0	0
		(74)	(All needlesticks combined)	(7)	(9.5)
Sodeyama et al, 1993	≥6	92	Needlestick	2[c]	2.2
Lanphear et al, 1994	≥5	50	Needlestick	3	6
		7	Sharp object injury	0	0
		7	Mucous membrane contamination	0	0
		4	Skin contamination	0	0
		1	Open wound contamination	0	0
		1	Human bite	0	0
		2	Unknown	0	0
		(72)	(All types combined)	(3)	(4.2)
Puro et al, 1995	≥6	331	Hollow-bore needlesticks	4	1.2
		105	Suture needle or sharp object injuries	0	0
		85	Mucous membrane contaminations	0	0
		125	Skin contaminations	0	0
		(646)	(All types combined)	(4)	(0.6)
Arai et al, 1996	≥12	56	All types combined	3	5.4
Takagi et al, 1998	≥6	251	All types combined	4	1.6
Hamid et al, 1999	6	53	Percutaneous (needlestick, sharp object injury)	2	3.8

[a] Data from studies with ≥50 accidents.
[b] 52 recipients were followed up for 12 months, 27 between 6 and 9 months, and 2 for 3 months.
[c] These cases had already been reported by Kiyosawa et al (1991).

screening tests for HCV, children with oncologic disease and those with heart surgery were at significant risk of acquiring HCV mainly by multiple transfusions. Follow-up studies showed anti-HCV-prevalence rates of 15% to 40% in these patients (Cesaro et al, 1997; Hoshiyama et al, 2000; Locasciulli et al, 1997; Vogt et al, 1999). Today, there is evidence that vertical transmission is overtaking transfusion as the principal course of HCV aquisition in children (Bortolotti et al, 1998), and the risk of perinatal transmission from mothers with detectable serum HCV-RNA may be as high as 5% to 10% (Giacchino et al, 1998; Ohto et al, 1994).

Clinical Course

Primary HCV infection in children varies from an acute active disease with elevated ALT to silent HCV-RNA appearance and anti-HCV seroconversion (Chang et al, 1994). The rate of progression to chronicity ranges from 60% to 85% (American Academy of Pediatrics, 1998; Hoshiyama et al, 2000; Resti et al, 1992). Spontaneous clearance of the virus has been reported in 11% to 29% of HCV-infected survivors of pediatric malignancies (Cesaro et al, 1997; Christensson et al, 2000; Locasciulli et al, 1997). In 67 children who were infected during cardiac surgery, the virus cleared in 45% after an average follow up of 20 years (Vogt et al, 1999).

The clinical course of hepatitis C is generally mild in children. No case of severe liver impairment was found in 67 patients cured of childhood leukemia over a period of 13 to 27 years (Locasciulli et al, 1997) or in 117 survivors of pediatric malignancy during a 14-year median follow up (Cesaro et al, 1997). Bortolotti et al (1994) reported only two cases of severe hepatitis with associated cirrhosis in 77 children after a mean observation period of 6 years. In 30 children who contracted their infection from mothers during birth, most showed biochemical features of liver damage in the first 12 months of life and progression to chronicity was observed in 80%, but associated liver disease was mild (Bortolotti et al, 1997). During the chronic phase of the disease, most children are asymptomatic although frequent fluctuations of viremia and ALT levels might occur (Ruiz-Moreno et al, 1999).

Histopathology

Histopathology of the liver in chronically infected children shows the typical characteristics of adult infection, such as lymphoid aggregate in the portal area, bile duct injury, and fatty changes. These changes and the extent of fibrosis tend to be mild (Hoshiyama et al, 2000; Kage et al, 1997). However, despite a mild degree of necroinflammatory activity, Badizadegan et al (1998) reported significant portal fibrosis in 23 of 40 children with chronic infection of relatively short duration. Garcia-Monzon et al (1998) compared 24 pediatric and 32 adult patients with similar mean duration of infection (11 $\pm$ 9 years) and with similar genotype distribution, and found lower ALT levels, lower viral load, and the mildest histological changes in children. Inui et al (1994) reported that the changes in liver histology in children with transfusion-associated chronic hepatitis C were more rapidly progressive in those with malignant disease or severe aplastic anemia than in patients without these diseases. A very poor long-term outcome was also observed in 4 children with primary hypogammaglobulinemia; 3 of them developed cirrhosis within 15 years after HCV infection (Bjoro et al, 1999).

Treatment

Interferon-α (IFN-α) and ribavirin are currently applied for the treatment of chronic HCV infection in adults. In children, there have been few studies of monotherapy with IFN-α; sustained response rates of 36% to 56% have been reported (Bortolotti et al, 1995; DiMarco et al, 1997; Fujisawa et al, 1995; Iorio et al, 1996; Matsuoka et al, 1997; Ruiz-Moreno et al, 1992; Sawada et al, 1998). Combination therapy with IFN-α and ribavirin has been found to significantly improve sustained response rates in adults (McHutchison et al, 1998). To our knowledge, there is only one report about a course of combined IFN-α and ribavirin in children; sustained virological response was observed in 6 (64%) of 11 patients after 48 weeks of combined therapy with IFN-α and ribavirin (Christensson et al, 2000). Currently, no vaccine is available, and the administration of immunoglobulin is not recommended (Centers for Disease Control and Prevention, 1998).

Discussion and Conclusion

We report an HCV outbreak in a pediatric oncology ward where the source of HCV infection and the mode of virus transmission could not be established. Similar nosocomial outbreaks with unknown virus transmission routes have been repeatedly documented; immunocompromised patients who had undergone many percutaneous procedures were often involved.

We can only speculate about the mechanisms involved in this outbreak. Patient P1 was the first identified case of HCV infection. However, the other patients showed no signs of acute hepatitis and were, at least in part, analyzed only retrospectively. Therefore, it was not possible to establish with certainty which patient was infected first. All patients were immunocompromised and had received blood products. Thus, the source of infection might have been a blood product with low-level HCV contamination. Minor and inadvertent violations of safety procedures, eg, when using multidose vials of saline to flush catheters, might have resulted in the spread of HCV. The virus might also have been transmitted from patient to patient by other known or unknown routes. For instance, patient P1 had the habit of biting his lip, and the virus might have spread from this bleeding wound.

Our findings emphasize that transmission of HCV poses a serious threat to patients and health care workers. Neither the magnitude nor the mechanisms involved in nosocomial HCV transmission are well understood yet. There is mounting evidence that only strict adherence to all possible infection control techniques and disinfection procedures can effectively prevent nosocomial transmission.

Materials and Methods

Serum samples were tested for HCV antibodies by the AxSYM third-generation microparticle enzyme immunoassay (EIA); positive results were confirmed with a semi-automated immunodot assay (Matrix HCV) (both from Abbott GmbH, Wiesbaden-Delkenheim, Germany). Detection of type-specific anti-HCV-antibodies (serotyping) was performed using the Murex HCV serotyping 1-6 assay (Murex Diagnostika GmbH, Burgwedel, Germany).

Serum samples were stored at $\leq -20°$ C, and RNA was isolated using the Qiamp Blood Kit (Qiagen, Hilden, Germany). HCV-RNA was detected by reverse transcription and nested PCR amplification with primers specific for the 5' noncoding region (Stuyver et al, 1993). Genotyping of these PCR products was performed with the commercial line probe assay Inno LiPA HCV II (Innogenetics, Ghent, Belgium).

For sequence analysis, the hypervariable region 1 (HVR1) of the E2/NS1 region of the HCV genome was reverse transcribed and amplified by nested PCR with primers derived from conserved flanking regions as described elsewhere (Booth et al, 1998). PCR products were purified with the QIA Quick Kit (Qiagen), and dye terminator cycle sequencing was carried out with a 373A ABI automated sequencer (PE Applied Biosystems Inc., Foster City, California). Sequence comparisons were performed with DISTANCES and GROWTREE of the Wisconsin Package, version 9.0 (Genetics Computer Group [GCG], Madison, Wisconsin) using the Kimura-2-parameter matrix and Neighbor joining.

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