Transfusion Medicine, 1998, **8**, 87–95

Hepatitis C in blood transfusion recipients identified at the Oxford Blood Centre in the national HCV look-back programme

A. E. Dike,* J. M. L. Christie,† J. B. Kurtz‡ and C.-G. Teo§ *NBS Oxford Blood Centre, †Department of Gastroenterology, John Radcliffe Hospital, Oxford, ‡Oxford Public Health Laboratory and §Central Public Health Laboratory, London, UK

Received 3 September 1997; accepted for publication 9 January 1998

SUMMARY. After the introduction in September 1991 of donor screening for hepatitis C, 95 potentially infectious blood donors who had given blood before this date were identified at the Oxford blood centre. Three hundred and ninety-nine blood components issued previously from these donors were identified in the course of the national HCV look-back programme. Of 399 questionnaires sent to hospital blood banks 392 were returned, identifying 290 recipients of whom 177 (61%) had died, and 113 (39%) were still alive 4–13 years after transfusion. One hundred and four recipients were traced and tested. Forty-nine recipients were not HCV infected. Forty-four of 58 (76%) recipients who received blood from donors found to be HCV RNA positive after September 1991 gave positive test results for HCV RNA. Eleven of 58 showed only antibody (anti-HCV), and 3/58 who had apparently received infectious blood showed no sign of past infection. The 11 who showed anti-HCV only, together with the three who showed no sign of past infection despite strong evidence of receiving HCV RNA-positive blood, had a mean age at transfusion of 27 years, compared with mean age at transfusion of 46 years in the 44 recipients with persistent HCV infection. Virus genotyping in 33/44 HCV RNA-positive recipients revealed five different genotypes. These did not seem to influence the outcome. Virus genotypes in 31 donor–recipient pairs showed complete concordance. Liver biopsies in 23/44 RNA-positive recipients showed minimal inflammation in four, mild in eight and moderate in 11. Liver fibrosis, Ishak grades 1–3, was present in 16/23 recipients. One other male recipient, not subjected to a liver biopsy, developed a hepatocellular carcinoma which caused his death at the age of 71, 8 years after transfusion.

Key words: recipient, HCV genotype, hepatitis C, liver biopsy, look-back.

Following the establishment of blood donor screening for hepatitis B surface antigen in the 1970s, it became apparent that other agents could cause post-transfusion hepatitis (Aach & Kahn, 1980). Hepatitis C virus (HCV) was first identified by Choo *et al.* (1989), and the development of tests to detect HCV antibody soon indicated that 90% of post-transfusion non-A, non-B hepatitis was caused by this newly discovered virus (Anon, 1989).

Early clinical studies of hepatitis C were based on patients with already detected liver disease, and therefore the overall risk of morbidity and mortality which included asymptomatic individuals was unknown. The introduction of blood donor screening for anti-HCV by the UK National Blood Service (NBS) in September 1991 led to the identification of many apparently healthy virus carriers, but once that testing was widely available it was realized that HCV infection, although mild to start with, could result in serious liver disease many years later. A retrospective study was carried out in Scotland (Ayob *et al.*, 1994) after the first 6 months of donor screening. Nine living recipients from 15 blood donors with chronic hepatitis C were tested, and all had signs of persisting infection. This pilot study indicated that substantial numbers of infected recipients would be identified in a nation-wide study. When a new antiviral drug, interferon-α, became available for the treatment of HCV infection the UK transfusion services decided that potentially infected recipients, i.e. those who had received blood in the past from donors now known to be positive for anti-HCV, should be traced so that those with chronic infection would have an opportunity for specific treatment.

The national HCV look-back programme was instituted by the UK Chief Medical Officer (Calman, 1995), on the recommendation of the Department of Health

Correspondence to: Dr A. E. Dike, Inverbroom, Manor Road, Towersey, Thame, Oxon OX9 3QS, UK.

Advisory Committee for the Microbiological Safety of Blood and Tissues for Transplantation (MSBT). The programme was directed and co-ordinated by the Medical Director of the National Blood Authority (Dr A. Robinson). The main aims of the study were to enable referral of HCV-infected patients to specialist care and to study a large number of people infected by blood transfusion from a known source at a known time, and thus to discover more about the natural history of hepatitis C. Results from the Oxford blood centre are reported here.

METHOD OF BLOOD DONOR SCREENING

Donor testing was carried out according to NBS Guidelines (1993) using second and third-generation enzyme immuno-assays (EIA) for hepatitis C antibody (anti-HCV), supplemented by recombinant immuno-blot assay (RIBA II/RIBA III) (Brown, 1995) for all EIA-positive samples.

In May 1995, at the beginning of the national HCV look-back study, the MSBT gave instructions for the combination of laboratory test results to be used to identify the blood donors whose previous donations were to be considered as potentially infectious for HCV. Donors were to be entered into the study if they had shown anti-HCV in two or more EIA tests using test kits from different manufacturers, together with a positive RIBA result. A year later the MSBT advised that blood donors whose sera were reactive in two or more EIAs but RIBA indeterminate (i.e. showing a strong reaction with only the NS4 (c33c) or core (c22) region on the immuno-blot strip (Wilbur, 1993) should also be considered as potentially infectious and should be included in the study. The reason for this was that several studies had shown that a small proportion of patients with indeterminate RIBA status were HCV RNA positive. Tests for HCV RNA were not routinely available in the early years of anti-HCV screening of blood donors, so it was realized that a number of blood donors with HCV RIBA indeterminate results were possibly HCV RNA positive and might have been infectious.

METHOD OF HCV LOOK-BACK

In March 1995 a standard set of questionnaire forms was issued by the National Blood Authority to all blood centres in England, Scotland, Wales and Northern Ireland together with guidance for counselling the recipients of potentially infectious blood components. Data to be gathered included whether transfusion of the blood component was documented in the hospital notes, and whether the recipient had other known risks for hepatitis C. Tests to be done on serum samples from the recipients, in designated virology laboratories, were as follows: two different EIA tests for anti-HCV, and a RIBA or similar supplementary test for the reactive samples. EIA-negative samples were to be tested by a reverse transcriptase polymerase chain reaction (PCR) to exclude rare cases which might be HCV RNA positive in the absence of detectable antibody. At Oxford, in addition to these minimum requirements, PCR tests were carried out on all the recipients with detectable anti-HCV, and on as many as possible of their linked donors, in many cases using samples which had been stored by the Oxford Public Health Laboratory (PHL) after finding them anti-HCV positive. HCV RNA was detected using an in-house reverse transcriptase PCR. Briefly, HCV RNA was extracted from 100 μL of serum using the method of Boom *et al.* (1990) and a nested PCR was carried out, using primers to the conserved 5′NTR region of the genome (Stuyver *et al.*, 1993). Reverse transcriptase (Advanced Biotechnologies, Epsom, Surrey, UK) was incorporated into the first round PCR mix. Following amplification, specific products were identified with reference to a molecular weight marker by agarose gel electrophoresis, stained with ethidium bromide and examined by UV transillumination.

Identification of the hospitals or other institutions to which blood had been issued was available from 1983 onwards in the Oxford blood centre computer database. Tracing of blood transfusion recipients was done with the co-operation of haematology departments in the 10 district hospitals, two Royal Air Force hospitals and five private hospitals served by the Oxford blood centre. Blood sampling of the traced patients was undertaken by hospital haematologists or other consultants or by family practitioners.

All blood transfusion recipients with evidence of hepatitis C infection were offered referral to the gastroenterology department of the John Radcliffe Hospital where investigations included serial liver function and PCR tests, and liver biopsies for those with detectable HCV RNA.

Where HCV RNA was detected in both donor and recipient linked pairs, HCV genotypes were determined at the Central Public Health Laboratory (CPHL) using a PCR-restriction fragment length polymorphism procedure. A 174-base-pair fragment of the 5′ noncoding region of the hepatitis C virus genome was amplified from serum RNA extracts. Primer sequences were those described by Lin *et al.* (1992). After PCR, aliquots of each product were digested with the restriction enzymes ScrfI, MvaI, HinfI and BstUI (Lin *et al.*, 1992) and the patterns were analysed after agarose gel electrophoresis followed by staining with ethidium bromide. This method is an adaptation of the one described by Pohjanpelto *et al.* (1995), and distinguishes between at least

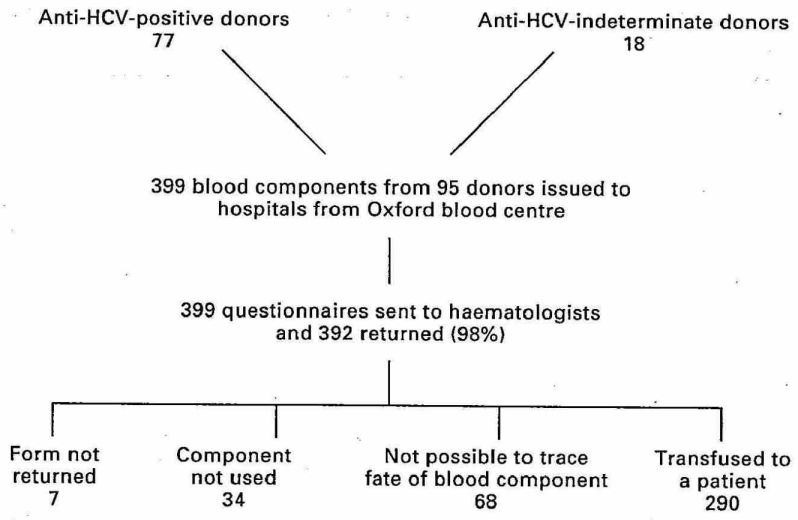

Fig. 1. Blood donors with previous donation records who were discovered to be anti-HCV positive or indeterminate by routine screening between 1 September 1991 and 1 February 1996.

nine different HCV subtypes (1a, 1b, 2a/c, 2b, 3a, 3b, 4, 5 and 6).

RESULTS

Donor identification (Fig. 1)

Seventy anti-HCV-positive donors with previous donations on record were identified in Oxford and seven more positive donors who had moved were identified in other blood centres and reported to Oxford, giving a total of 77 anti-HCV-positive donors for the look-back. In addition, 14 anti-HCV indeterminate donors with previous donations were entered into the look-back together with four more reported to Oxford from other centres (adding 18).

Look-back results (Figs 1 and 2)

Three hundred and ninety-nine potentially infectious blood components from the 95 identified donors had been issued for clinical use, and questionnaires relating to each of these blood components were sent to the hospital haematologists (45 of these were sent to hospitals via four other blood centres which had received blood components from Oxford, and results were returned to Oxford). The questionnaires had been designed to ascertain the identity of the recipients, and then to examine hospital notes to find out whether the recipients might be alive and traceable. Of the 399 questionnaires 392 (98.2%) were returned. These showed that 34 blood components had not been transfused. Sixty-eight (19%) of the remaining 358 components had not been traced for the following reasons: 37 were unrecorded in the hospital transfusion laboratories and 22 had insufficient patient details recorded to allow identification of the hospital records; there were six cases with adequate details recorded in the hospital blood bank but with no traceable hospital records, and three identified patients had hospital records but were not traceable by the Family Health Service Authorities.

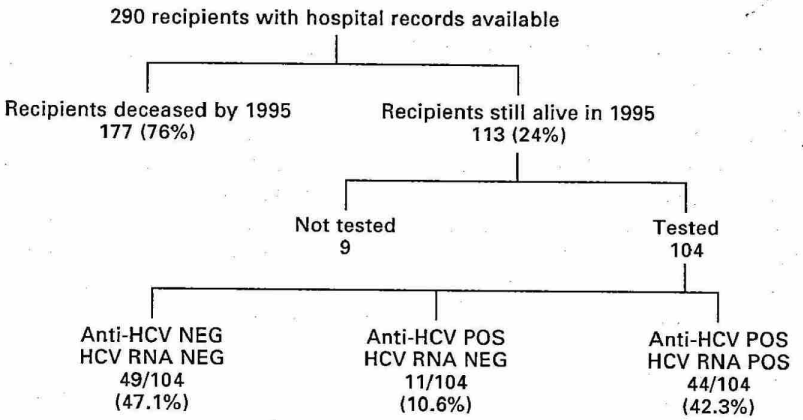

Fig. 2. The outcomes for 290 transfused recipients who were traced via hospitals and family practitioners.

Table 1. Forty-nine recipients who were anti-HCV negative and HCV RNA negative in 1995–1996 following blood transfusions received 1983–1993.

Recipient ID	Sex	m/yr transfused	Age transfused	Transfusion confirmed[1]	Donor-ID	Donor's anti-HCV[2]	Donor's HCV RNA[3]	Donor's infection date[4]
105	M	10/83	31	Yes	7	Pos	Pos	NK
*57	M	06/86	18	Yes	9	Pos	Pos	NK
17	M	08/84	27	Yes	10	Pos	Pos	1989
41	F	02/86	25	No	10			
81	M	11/87	64	Yes	11	Pos	Pos	1988
9	F	06/85	53	No	11			
75	F	02/85	61	Yes	13	Pos	Pos	1989
90	F	06/91	35	Yes	14	Pos	Pos	1983
111	F	05/86	75	Yes	14			
53	F	06/87	62	Yes	15	Pos	Pos	1990
*52	F	03/90	33	Yes	18	Pos	Pos	1978
116	M	06/91	27	No	25	Pos	Pos	NK
4	M	07/91	73	Yes	30	Ind	Neg	1984
86	M	01/88	55	Yes	30			
68	M	11/85	18 mths	No	30			
67	F	10/83	54	Yes	50	Pos	Pos	NK
46	F	10/89	40	Yes	54	Pos	Pos	1985
2	F	01/83	25	No	57	Pos	Pos	1965
*49	F	01/84	9	Yes	57			
33	M	02/90	28	Yes	66	Ind	Neg	NK
59	F	02/89	24	Yes	66			
21	M	08/91	18 mths	No	66			
14	M	02/90	2 mths	No	66			
55	M	03/90	62	No	66			
29	M	08/90	12	Yes	66			
54	M	09/86	54	Yes	76	Ind	Neg	NK
94	M	03/87	2 mths	Yes	77	Ind	Neg	NK
5	M	10/89	43	No	77			
79	M	03/89	5 days	Yes	77			

Two hundred and ninety recipients were identified, of whom 177 (61%) had died. Of the remaining 113 patients who were alive and traceable, there were nine who were either very old or terminally ill or mentally incapable of understanding the reasons for blood testing and their doctors considered that they should not be approached. Close relatives or carers were informed of the possibility of hepatitis C in these patients. This left 104 recipients who could be counselled and who agreed to be tested and on whom the following results are based. They were linked to 43 of the 77 anti-HCV-positive blood donors and to 11 of the 18 anti-HCV-indeterminate donors. No living recipients were traced to the other 41 potentially infectious donors.

The 104 traced and tested recipients were divided into three categories: (a) anti-HCV and HCV RNA both negative; (b) anti-HCV positive and HCV RNA negative; (c) anti-HCV and HCV RNA both positive.

Recipient anti-HCV and HCV RNA negative (Table 1)

Forty-nine recipients (47%) were shown to be anti-HCV negative and had no detectable HCV RNA. These recipients were transfused between 1983 and 1993, and were tested in 1995 and 1996. They could be subdivided into three categories according to the status of 28 implicated donors, as follows.

(i) Ten recipients were transfused from seven donors, all of whom were documented as having acquired hepatitis C infections some time later than these traced donations: three regular blood donors who were anti-HCV negative between 1991 and 1993 showed seroconversion between

Table 1. *Continued*

Recipient ID	Sex	m/yr transfused	Age transfused	Transfusion confirmed[1]	Donor-ID	Donor's anti-HCV[2]	Donor's HCV RNA[3]	Donor's infection date[4]
3	F	05/91	59	No	77			
23	M	03/85	12 days	Yes	77			
38	M	04/85	73	Yes	77	Ind	Neg	NK
24	F	09/89	9	No	77			
45	F	03/87	10	No	77			
22	F	07/90	75	Yes	78	Ind	Neg	NK
50	M	01/86	54	Yes	79	Ind	Neg	NK
108	F	02/91	78	Yes	79			
16	M	10/83	30	Yes	80	Ind	Neg	NK
104	M	02/83	64	No	84	Pos	Neg	NK
51	M	06/89	77	Yes	84			
101	M	01/92	54	Yes	94	Pos	Pos	1992
36	M	03/93	70	Yes	95	Pos	Pos	1993
92	F	10/92	53	Yes	95			
87	M	02/93	14	Yes	129	Pos	Pos	1994
112	M	02/89	11 days	Yes	133	Ind	NT	NK
39	M	07/88	58	Yes	135	Ind	NT	NK
31	M	02/88	44	No	137	Ind	NT	NK
8	M	09/90	78	Yes	145	Pos	Neg	NK
91	M	10/86	45	Yes	146	Ind	Neg	NK

Each recipient and donor has an Oxford identification number (ID). Recipients are grouped in each table with their linked donors, who are listed in numerical order. Note that some donors appear in more than one of the tables and that some donors transmitted HCV infection to several recipients.

1. Recipient's notes do not confirm that the blood component was transfused. 2. Anti-HCV tests in the donor after September 1991 were positive or indeterminate as defined in text. 3. HCV RNA tests in the donor after September 1991 were positive or negative. NT = not tested. 4. Year of infection of the donor if known or highly probable. NK = not known. 5. HCV genotypes in donors and recipients if tested. (Samples containing detectable HCV RNA were not available for all subjects.). 6. Liver function: ALT and/or AST within laboratory's normal range, or at least 50% above upper limit of range, or results fluctuating. 7. Symptoms elicited when patient first assessed. 8. Histological description of the degree of inflammation in liver biopsy. 9. Inflammation score (see text). 10. Histological description of the degree of fibrosis in liver biopsy. 11. Fibrosis score (see text). (Notes 5–11 apply to Tables 2 and 3.)

1994 and 1996; three other donors acquired hepatitis C at later dates, from blood transfusions they themselves received, and were discovered as 'look-back recipients'; and one donor admitted injecting himself with drugs for a short period after the donation under investigation. Later recipients of two of the seroconverted donors were infected (see Recipient anti-HCV and HCV RNA positive), but the other five donors had no later recipients available for testing.

(ii) Twenty-nine recipients with no evidence of infection were transfused from 13 donors, 11 of whom had indeterminate antibody status by RIBA as defined above, and two of whom had positive HCV antibody results by all tests. However, all these donors were HCV RNA negative by PCR on several occasions, and none of them was linked to any infected recipients. Transfusion of the traced blood components was specifically recorded in the hospital notes for only 19 of these 29 recipients, but transfusions from 12 of the 13 donors were recorded in the notes of one or more of their recipients.

(iii) Ten recipients with no evidence of infection received blood from seven donors who were anti-HCV and HCV RNA positive when screened after 1991. One of the seven donors (donor-ID 9) was linked to two other infected recipients transfused before and after the date of this traced donation; another donor (donor-ID 18) had transmitted hepatitis C infection to a second recipient of the same donation; and a third donor (Donor-ID 57) had transmitted HCV 6 months earlier to another recipient. Since in the three recipients (ID numbers 49, 52, 57 marked with asterisks in Table 1) there was clear evidence in the hospital notes that the transfusions had been given, this suggests that they had either resisted or recovered from infection, retaining no detectable antibody; they were one female aged 9, one female aged 33

Table 2. Eleven recipients who were anti-HCV but HCV RNA negative in 1995–1996 following blood transfusions received 1987–1991.

Recipient ID	Sex	m/yr transfused	Age transfused	Donor-ID	Donor's anti-HCV[2]	Donor's HCV RNA[3]	HCV genotype of donor[5]
82	F	12/90	23	16	Pos	Pos	1a
93	M	08/91	3 days	18	Pos	Pos	3a
56	F	02/90	2 days	18			
27	F	08/89	55	53	Pos	Pos	2b
11	F	10/87	39	64	Pos	Pos	5
89	M	03/90	7	65	Pos	Pos	1a
80	F	08/91	62	72	Pos	NT	NT
115	F	06/91	29	114	Pos	Pos	NT
95	M	07/86	30	128	Pos	Pos	1b
65	F	10/87	30	128			
110	F	12/90	25	144	Pos	Pos	2b

and one male aged 18 when transfused in 1984, 1990 and 1986, respectively. Further details are presented in Table 1 and their respective donors can also be seen in Tables 2 and 3 linked to infected recipients.

Recipient anti-HCV positive and HCV RNA negative (Table 2)

Eleven recipients, found to have anti-HCV only, were transfused from nine donors who had evidence of chronic hepatitis C infection. These recipients have been referred to liver specialists and have been retested several times over a period of 6–18 months, showing consistently normal liver transaminases and negative PCR results. None produced symptoms of liver disease. No liver biopsies have been carried out in this group of patients, but they will continue to be followed up in order to confirm that their PCR tests remain negative. Seven out of the nine donors for this group are linked to other recipients who were found to be HCV RNA positive, as can be seen in Table 3.

Recipient anti-HCV and HCV RNA positive (Table 3)

Forty-four recipients in this category were transfused from 29 donors. This includes 42 recipients transfused with blood from 27 donors identified as seropositive, and two recipients transfused in 1992 and 1993 from two blood donors (donor-IDs 94 and 95) whose blood had given clearly negative results in routine hepatitis C screening tests. A subsequent donation from each of the two donors was positive for anti-HCV and HCV RNA, showing that these two donors were almost certainly viraemic in 1992 and 1993, but had donated blood before anti-HCV was detectable. HCV genotyping in both donor–recipient pairs showed matching virus types.

HCV genotyping in 25 donors showed a variety of genotypes (six with type 1a, five with 1b, three with 2b, 10 with 3a and one with type 5). Thirty-three recipient samples were tested and all the HCV genotypes seen in the donors were also found in the recipients. There were eight with type 1a, five with 1b, three with 2b, 16 with 3a and one with type 5. Thirty-one donor–recipient pairs showed matching HCV genotypes, providing supporting evidence of transfusion-transmitted infection. There were no genotype mismatches in the donor–recipient pairs.

Of the 44 anti-HCV- and HCV RNA-positive recipients, 28 have been investigated in the Oxford gastroenterology department and four are being seen by liver specialists in other hospitals. One recipient, a 23-year-old asymptomatic female, has not yet agreed to a referral. Eleven recipients discovered to be infected with HCV have not been referred to gastroenterologists either because they are elderly and asymptomatic or because they already have serious medical conditions other than liver disease. Most of the referred patients were asymptomatic, but four complained of lethargy, one of nausea and one had symptoms and signs consistent with cirrhosis (ascites and encephalopathy). This male patient, who was aged 63 when transfused in 1989, developed a hepatocellular carcinoma (HCC) and died in 1997. In this group of patients transaminase results were repeatedly within the laboratory's normal range in 21/44 (48%), 50% or more above the upper limit of normal in 17/44 (39%) and fluctuating in 6/44 (13%). Liver biopsy was recommended to most of the referred patients with evidence of continuing viraemia (PCR positive on two

Table 3. Forty-four recipients who were anti-HCV and HCV RNA positive in 1995–1996 following blood transfusions received 1989–1993.

Recipient ID	Sex	m/yr transfused	Age transfused	LFT[6]	Symptoms[7]	Inflammation[8]	Inf-score[9]	Fibrosis[10]	Fib-score[11]	Donor-ID	Donor's anti-HCV[2]	Donor's HCV RNA[3]	HCV genotype[5] Donor	HCV genotype[5] Recipient
63	F	04/84	36	Normal	None	Mild	2	None	0	8				1a
99	M	05/87	55	Abnormal	None	Mild	2	Mild	1	8	Pos	Pos	1a	NT
32	M	05/88	61	Normal	None	Moderate	4	Moderate	3	8				1a
100	F	06/84	54	Normal	None	Mild	2	Mild	1	9	Pos	Pos	1b	1b
84	F	12/90	34	Abnormal	None	Moderate	5	Mild	1	9				1b
83	F	06/91	36	Abnormal	Lethargy	Mild	2	Moderate	2	16	Pos	Pos	1a	1a
42	F	01/91	26	Normal	None	Minimal	2	None	0	17	Pos	Pos	1b	1b
26	F	09/91	80	Normal	None	Not referred				18				3a
69	M	04/91	33	Abnormal	None	Refused biopsy				18	Pos	Pos	3a	3a
25	M	09/90	40	Abnormal	None	Mild	2	Mild	1	18				NT
6	M	06/89	25	Normal	None	Not referred				19	Pos	NT	NT	NT
12	M	02/91	68	Normal	None	No biopsy (pcr now neg)				20	Pos	Pos	3a	3a
62	F	04/90	27	Normal	None	Minimal	1	None	0	22	Pos	Pos	3a	3a
34	M	04/91	40	Abnormal	Nausea	Moderate	6	Moderate	2	22				3a
15	F	12/90	55	Abnormal	None	No biopsy (on Warfarin)				24	Pos	Neg 1996	NT	3a
103	M	05/91	56	Abnormal	Lethargy	Moderate	5	Moderate	2	24				3a
107	F	08/89	27	Fluctuate	None	Moderate	4	Moderate	2	27	Pos	Pos	1b	1b
58	F	06/91	69	Normal	None	Not referred				31	Pos	Pos	3a	NT
13	F	08/89	31	Normal	None	Minimal	1	None	0	50	Pos	Pos	NT	NT
10	M	04/89	63	Abnormal	Ascites	No biopsy		HCC		51	Pos	Pos	NT	NT
74	F	05/88	29	Fluctuate	None	Mild	2	Mild	1	53	Pos	Pos	2b	2b
1	F	06/85	45	Normal	NK	Unrelated death 1996				55	Pos	Pos	3a	3a
47	M	05/88	67	Normal	None	Not referred				55				3a
76	M	10/89	76	Abnormal	NK	Not referred				56	Pos	Pos	3a	3a
64	F	07/83	54	Normal	None	Moderate	4	Moderate	2	57	Pos	Pos	1a	1a
18	M	02/87	68	Abnormal	NK	Not referred				60	Pos	Pos	1a	1a
19	F	09/87	44	Fluctuate	Lethargy	Mild	1	Mild	1	61				3a
98	F	07/88	44	Fluctuate	None	Minimal	2	None	0	61	Pos	Pos	3a	3a
40	M	07/87	47	Normal	NK	Not referred (dialysis)				61				3a
73	M	02/89	44	Abnormal	None	Moderate	5	Moderate	2	63	Pos	NT	NT	NT
66	M	07/91	83	Abnormal	NK	Not referred				64	Pos	Pos	5	5
7	M	03/90	6	Normal	None	Under review				65				1a
20	F	04/89	18	Normal	None	No biopsy				65	Pos	Pos	1a	NT
43	M	09/88	55	Abnormal	NK	Not referred				65				1a
114	F	10/90	40	Normal	None	No biopsy				65				NT
35	F	08/91	23	Normal	None	Moderate	4	None	0	73	Pos	Pos	3a	3a
97	F	12/92	44	Abnormal	None	Moderate	4	None	0	94	Pos	Pos	1a	1a
71	F	09/93	82	Normal	NK	Not referred				95	Pos	Pos	2b	2b
88	F	03/89	18	Normal	None	Not referred				103	Pos	Pos	1b	1b
72	F	02/87	66	Normal	NK	Not referred				103				NT
113	F	02/87	42	Abnormal	Lethargy	Mild	2	Mild	1	112	Pos	Pos	NT	NT
30	M	09/90	53	Abnormal	NK	Not referred				143	Pos	Pos	3a	3a
48	M	01/89	47	Fluctuate	None	Moderate	4	Moderate	3	143				3a
102	M	06/91	33	Fluctuate	None	Moderate	5	Mild	1	144	Pos	Pos	2b	2b

or more occasions). One patient became PCR negative when retested in the clinic, one patient refused to have a liver biopsy and in two patients liver biopsy was contra-indicated by abnormal blood coagulation. Three others have not yet had liver biopsies.

Liver biopsy results in 23 patients were classified by the system proposed by Ishak *et al.* (1995), giving numerical scores for the degree of liver inflammation (range 0–18) and the degree of fibrosis (range 0–6). Seven recipients, all female, with a mean age of 33 years when transfused 4–12 years earlier, showed no liver fibrosis. Sixteen recipients, eight male and eight female, showed mild to moderate liver fibrosis. The mean age of these 16, together with the man with HCC, was 46 years when transfused 5–13 years earlier. The clinical findings and liver biopsy results are summarized in Table 3.

DISCUSSION

The look-back exercise has identified a number of mainly asymptomatic blood transfusion recipients who were infected with hepatitis C at a known time, making them an ideal group of all ages in whom to study the natural history of this infection. The pilot study by Ayob *et al.* (1994) predicted that it might be possible to find approximately one infected recipient per donor. At Oxford 44 recipients with chronic infection were found and were linked to 43 of the 77 antibody-positive donors. Twenty-eight recipients and 22 of the linked donors and 70 other former blood donors with hepatitis C infection are now being followed up by Oxford gastroenterologists; others are being managed elsewhere. Unless there are contra-indications, most persistently viraemic blood transfusion recipients with any histological evidence of liver disease are being offered treatment with specific antiviral drugs. The ultimate benefit of treatment is unknown and the patients are encouraged to share in decision-making about their management. This is similar to that suggested by Foster *et al.* (1997).

Thirty-nine of the 104 tested recipients had probably not received infectious blood, because 13 of the linked donors showed only anti-HCV and no HCV RNA, and seven more donors were known to have been infected at later dates. Ten recipients, transfused from seven other donors who were anti-HCV and HCV RNA positive, showed no sign of infection; some may have recovered, and some may not have received infectious blood; for example, the donors might have acquired their infections at later dates, or the transfusions might not have been given since in four cases confirmation of transfusion was not found in the hospital notes. The potential infectivity of donor blood collected between 1983 and August 1991 could not be tested directly as no donor serum samples had been stored during that period. The Oxford study revealed no infected recipients from 11 anti-HCV-indeterminate donors, but the numbers are small and accumulated figures from the national study will probably show that a few currently RIBA-indeterminate donors did transmit HCV. In most centres, as at Oxford, it will probably not be possible to re-test the relevant blood donations from archived samples. It is now known that some HCV infections resolve spontaneously, and that test results for hepatitis C may become weaker or disappear with time. Therefore an attempt to trace all potentially infected recipients must include donors with currently indeterminate RIBA results as defined above. The Oxford study showed no recipients with detectable HCV RNA when anti-HCV tests were negative. This would possibly occur only rarely in immunodeficient patients; the combined national results will show if any such cases have been detected.

A recent report on needle-stick injuries by Dore, Kaldor and McCaughan showed that people who were HCV RNA positive transmitted infection, whereas HCV RNA-negative people did not. Likewise in the Oxford study it was shown that 35 HCV RNA-positive donors transmitted infection in 55/58 cases (95%). Of the 58 infected recipients 44 (76%) were persistently HCV RNA positive after intervals of 4–13 years. Eleven other recipients were in good health and had indications of recovery from infection since they were repeatedly negative for HCV RNA although anti-HCV positive. There were three recipients with no detectable anti-HCV or HCV RNA after 7–13 years in spite of strong evidence of transfusion with infectious blood. These 14 people, four of whom were young children, had a mean age of 27 years when transfused and a sex ratio of four males to 10 females (Tables 1 and 2). By comparison, the 44 recipients with chronic hepatitis C infection had mean age at transfusion of 46 years, with 10 recipients over the age of 60 and the sex ratio in this group was 20 males to 24 females (Table 3). Logistic regression analysis indicates that the age difference in the two groups did not quite achieve statistical significance ($0.05 < P < 0.1$), and that numbers were too small to deduce any influence of sex on the outcome of infection. However, the findings accord with a recent report by Poynard *et al.* (1997) who found that age at infection together with male sex influenced the outcome more than differences in virus genotypes. In our study, as in theirs, liver biopsies in the younger patients showed less fibrosis. Also as found by Poynard, we found a variety of HCV genotypes in the recipients with chronic infection and the same range of genotypes was present in donors linked to recipients who had recovered. This gives no indication of a more severe effect due to a particular virus genotype.

HCV is known to lead to liver fibrosis and hepatocellular carcinoma, but progression to serious liver disease

is extremely variable. Results from the Oxford part of the national HCV look-back study showed that 14/58 (24%) of those infected have probably made a full recovery from hepatitis C, but that 44/58 (76%) have become chronic virus carriers and 17 of these had histological evidence of liver disease 4–13 years from the date of infection.

It is known that hepatitis C can lead to liver failure after many years, so the younger patients detected by the national look-back study will benefit if the virus can be eliminated before serious liver disease occurs. The effects of treating these patients are currently being studied in many specialist liver units. Results from the national HCV look-back study will provide valuable information about factors that influence the outcome of HCV infection acquired at a known point in time. Moreover, many transfusion recipients and former blood donors with chronic hepatitis C infection will now, as a result of the look-back programme, receive long-term specialist management which will allow early diagnosis of malignancy and, for some of the younger blood transfusion recipients, may prevent the risk of liver failure in middle age.

ACKNOWLEDGMENTS

We thank Dr Roger Chapman, Consultant Gastoenterologist at Oxford, for clinical information about his patients, and we thank the liver specialists and virologists in other parts of the country who have contributed data for this report; also Joan Healey (Secretary, Oxford PHL Virology) for her assistance with finding samples and reporting results; Tracey Stone, Research Nurse at Oxford 1995–6; Leicester Gill (Computer and Information Scientist, University of Oxford, Institute of Health Sciences) for statistical advice; Kathryn Harris, CPHL, for HCV genotyping; and Oxford Medical Illustration for the artwork.

REFERENCES

Aach, R.D. & Kahn, R.A. (1980) Post-transfusion hepatitis: current perspectives. *Annals of Internal Medicine*, **92,** 539–546.

Anon (1989) *Proceedings of the First International Symposium, Hepatitis C Virus, Rome, Italy.*

Ayob, Y., Davidson, J.I., Baxter, A., Jordan, A., Yap, P.L. & Gillon, J. (1994) Risk of hepatitis C in patients who received blood from donors subsequently shown to be carriers of hepatitis C virus. *Transfusion Medicine*, **4,** 269–272.

Boom, R., Sal, C.J.A., Salimans, M.M., Jansen, C.L., van Dillen, W. & van der Noorda, J. (1990) Rapid and simple method for purification of nucleic acids. *Journal of Clinical Microbiology*, **28,** 495–503.

Brown, J.L. (1995) Diagnosis of hepatitis C virus infection. *Hospital Update Supplement on Current Aspects of Chronic Hepatitis C*, 11–12.

Calman, K.C. & Hepatitis, C.&. (1995) Blood Transfusion Look-Back. *CMO Letter*, **CMO (95) 1.**

Choo, Q.L., Kuo, G., Weiner, A., Overby, L.R., Bradley, D.W. & Houghton, M. (1989) Isolation of a c-DNA clone derived from a blood-borne viral hepatitis genome. *Science*, **244,** 359–362.

Dore, G.J., Kaldor, J.M. & McCaughan, G.W. (1997) Systematic review of polymerase chain reaction in defining infectiousness among people infected with hepatitis C virus. *British Medical Journal*, **315,** 333–337.

Foster, G.R., Goldin, R.D., Main, J., Murray-Lyon, I., Hargreaves, S. & Thomas, H.C. (1997) Management of chronic hepatitis C: clinical audit of biopsy based management algorithm. *British Medical Journal*, **315,** 453–458.

Guidelines for the Blood Transfusion Service (1993) *Section 1, Annex 3*. HMSO, London.

Ishak, K., Baptista, A., Bianchi, L., Callea, F., *et al.* (1995) Histological grading and staging of chronic hepatitis. *Journal of Hepatology*, **22,** 696–699.

Lin, H.J., Shi, N., Mizokami, M. & Hollinger, F.B. (1992) Polymerase chain reaction assay for hepatitis C using a single tube for reverse transcripion. *Journal of Medical Virology*, **38,** 220–225.

Pohjanpelto, P., Widell, A., Lappalainen, M. & Asikainen, K. (1995) *Progress in Clinical Virology.* Abstracts of Joint Meeting between European Group for Rapid Viral Diagnosis, and European Society against Virus Diseases, Prague, 10–14 September, 52.

Poynard, T., Bedossa, P. & Opolon, P. (1997) Natural history of liver fibrosis progression in patients with chronic hepatits C. *Lancet*, **349,** 825–832.

Stuyver, L., Rossau, R., Wyseur, A., Duhamel, M., Vanderbroght, B., van Heuverswyn, H. & Maertens, G. (1993) Typing of hepatitis C virus isolates and characterisation of new sub-types, using a line probe assay. *Journal of General Virology*, **74,** 1093–1102.

Wilber, J.C. (1993) Development and use of laboratory tests for hepatitis C infection: a review. *Journal of Clinical Immunoassay*, **16,** 204–207.