Canada has not introduced the non-A, non-B (NANB) surrogate tests respectively.

Transfusion hepatitis rates were 8.6 and 6.8 per 1000 respectively (p=0.06). After withholding with 5.0 in the withhold group (p=0.05), and the HCV recipients was 20.2 in the no-withhold group compared post-transfusion hepatitis rate per 1000 transfusion was screened for anti-HCV. During this time the overall hepatitis C virus after transfusion before all donor blood NANB surrogate testing was due to reduced frequency of hepatitis C rate by 70% (p=0.05). Most of the benefit of transfusion hepatitis by 40% (p=0.065) and the NANB surrogate positive units reduced the overall post-transfusion hepatitis due to hepatitis A, B, C, non ABC, Epstein-Barr virus (EBV) and cytomegalovirus (CMV). Withholding of blood containing NANB surrogate positive units reduced the overall post-transfusion hepatitis rate by 40% (p=0.065) and the hepatitis C rate by 70% (p=0.05). Most of the benefit of NANB surrogate testing was due to reduced frequency of hepatitis C virus after transfusion before all donor blood was screened for anti-HCV. During this time the overall post-transfusion hepatitis rate per 1000 transfusion recipients was 20.2 in the no-withhold group compared with 5.0 in the withhold group (p=0.05), and the HCV hepatitis rate was 12.6 and 6.0 respectively (p=0.06). After the introduction of HCV screening, the overall post-transfusion hepatitis rates were 8.6 and 6.8 per 1000 (p=0.05) respectively.

Our study indicates that screening of blood donors with the NANB surrogate markers was of value in reducing HCV infection before HCV screening began, but subsequently the value of screening cannot be clearly established.

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Summary
Canada has not introduced the non-A, non-B (NANB) surrogate marker tests (antibodies to hepatitis B core antigen and alanine aminotransferase) to screen donated blood. We evaluated the effect of NANB surrogate markers in reducing post-transfusion hepatitis in a prospective randomised intervention study.

From 1988 to 1992, 4588 subjects were enrolled into two study groups that received allogeneic blood from which units positive for NANB surrogate markers were either withheld (n=2311) or not withheld (n=2277). We also assessed a simultaneous non-randomised cohort (n=650) of subjects who received only syngeneic blood. All subjects were followed up for 6 months and assessed for the presence of post-transfusion hepatitis due to hepatitis A, B, C, non ABC, Epstein-Barr virus (EBV) and cytomegalovirus (CMV). Withholding of blood containing NANB surrogate positive units reduced the overall post-transfusion hepatitis rate by 40% (p=0.065) and the hepatitis C rate by 70% (p=0.05). Most of the benefit of NANB surrogate testing was due to reduced frequency of hepatitis C virus after transfusion before all donor blood was screened for anti-HCV. During this time the overall post-transfusion hepatitis rate per 1000 transfusion recipients was 20.2 in the no-withhold group compared with 5.0 in the withhold group (p=0.05), and the HCV hepatitis rate was 12.6 and 6.0 respectively (p=0.06). After the introduction of HCV screening, the overall post-transfusion hepatitis rates were 8.6 and 6.8 per 1000 (p=0.05) respectively.

Our study indicates that screening of blood donors with the NANB surrogate markers was of value in reducing HCV infection before HCV screening began, but subsequently the value of screening cannot be clearly established.

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Introduction
A prospective study of post-transfusion hepatitis in Canada in 1984–85 showed an overall post-transfusion hepatitis frequency of 92 per 1000 allogeneic blood recipients, with a post-transfusion frequency of hepatitis C (HCV) of 31 per 1000 recipients. Since 1985, many measures were introduced by blood-collection agencies worldwide to try to improve the safety of the blood supply. These included the introduction of screening for HIV-1, human T-cell lymphotropic virus (HTLV) type I, HBsAg, and direct questioning of blood donors about relevant medical information and lifestyle. In 1986, agencies in the USA introduced two tests to screen blood donors, to reduce the frequency of non-A, non-B (NANB) post-transfusion hepatitis. Thus, donors with an increased concentration of the hepatic enzyme alanine aminotransferase (ALT) or the presence of antibodies to the hepatitis B core antigen (anti-HBc) were excluded from donating blood. These two NANB surrogate markers were used because of results from two prospective studies of allogeneic blood product recipients in the 1970s. This decision was made without the benefit of data from prospective intervention studies showing efficacy.

Because of the lack of such evidence, the Canadian Red Cross Society and some blood transfusion services in western Europe did not screen blood donors for NANB surrogate markers. We thought a randomised double-blind trial was needed in Canada to assess the frequency of post-transfusion hepatitis and so see whether the withholding of donor blood positive for the NANB surrogate markers would reduce the frequency of post-transfusion hepatitis.

While our study was in progress the genome of HCV was elucidated.1 Testing blood donors for antibodies to HCV was introduced in Canada in May, 1990. Subjects were involved in our study before and after the introduction of anti-HCV testing.

Patients and methods

Patients
Participating institutions included 3 Canadian Red Cross Society Blood Centres (Hamilton, Toronto, and Winnipeg), and 13 university-affiliated hospitals (6 with McMaster University, 5 with the University of Toronto, and 2 with the University of Manitoba). Consecutive adult patients who required transfusion (red blood cells and/or plasma) and were admitted to participating hospitals, were screened for eligibility by trained research personnel before transfusion. We obtained written informed consent from each subject before study entry. Specifically, we told potential subjects that previous results suggested that NANB surrogate tests could predict whether blood donors carry viruses that could cause post-transfusion hepatitis. Exclusion criteria were: the presence of red cell alloantigens that would have made the provision of blood products difficult; chronic liver disease; known alcohol abuse;
blood product transfusion in the past 6 months; presence of malignant disease with metastases; a medical condition with a life expectancy of less than 9 months; and a pre-surgery requirement for informed consent, eligibility, and follow-up procedures. We thus had three study groups.

Laboratory methods

All blood samples were tested in a central laboratory at the liver study unit at Mount Sinai Hospital. We measured alanine aminotransferase concentration. Baseline data for each subject included relevant demographic data; the type and number of blood products transfused; the type of surgery; medications; and perioperative hypotensive episodes. Our protocol specified that all subjects would be followed up for 6 months after blood transfusion with visits at 3, 6, 8, 12, and 24 weeks. At each visit, we took a blood sample for alanine aminotransferase measurement and asked the patient to fill in a short questionnaire. We stored each sample taken for alanine aminotransferase in aliquots at -70°C. All blood products we used on our subjects were tested and were negative for: VDRL, HBsAg, HIV-1 antibodies, HTLV-2 antibodies, and from May 1, 1990, the first generation anti-HCV test.

To determine the background frequency of post-transfusion hepatitis, we also studied a simultaneous cohort of 650 non-randomised control subjects having surgical procedures but receiving only autologous blood. This group had the same requirements for informed consent, eligibility, and follow-up procedures. We thus had three study groups.

Classification of post-transfusion hepatitis events

Subjects were classified as having post-transfusion hepatitis when: the serum alanine aminotransferase concentrations were increased to at least 2-5 times the upper limit of normal; the alanine aminotransferase of a second serum sample was increased to at least twice the upper limit of normal within 7-10 days of the initial increase; and other potential causes of abnormal liver function were excluded. The final post-transfusion hepatitis diagnosis of each subject was made by an events committee, consisting of an infectious disease specialist and two hepatologists. The committee was not involved in any other part of the study and was unaware of the transfusion group when reviewing data. We based the aetiological diagnosis of post-transfusion hepatitis caused by either HBV or HCV on reviewing data. We based the aetiological diagnosis of post-transfusion hepatitis caused by either HBV or HCV on reviewing data.
analyses had not been specified in the original design. We 13
with covariates for group and time, and with and without testing for each time-period separately and for the secondary Breslow-Day test. Homogeneity of odds ratios among the strata was assessed by the Mantel-Haenszel analysis stratified by centre and by time. The frequency of post-transfusion hepatitis in the withhold group. Because of the small numbers of events for the secondary outcomes by time-periods, we also did exact logistic regression analyses. To increase the low power of the standard logistic regression test for differential surrogate testing effects between the two time periods we pooled the data from the withhold groups for blood donated before and after HCV screening. We estimated the relative risks associated with transfusion of a marker-positive unit in each period for each of the outcomes with logistic regression disregarding odds ratios. Indicator covariates for the type of donor units received (any units that were positive for HCV antibodies or any units with alanine aminotransferase units above 40) were used to investigate the association with each marker on its own and in combination. Tests for differential effects by time were done by including interactions between time-period and unit type. The associations of post-transfusion hepatitis with the total number of units transfused and with the numbers of negative and positive units transfused were investigated by logistic regression. We used Poisson regression to estimate per unit rates and CIs with an offset for the numbers of units transfused.

Results
Frequency of post-transfusion hepatitis
Table 1 shows the demographic and other characteristics of the study subjects in the two allogeneic transfusion groups. The participants in the study groups were generally similar in sociodemographic, and transfusion characteristics. About 15% of subjects received non-study or misallocated blood. Exclusion of these cases from the analysis did not change the study findings. The non-randomised cohort of recipients of syngeneic blood differed in several respects from those in the non-withhold and the withhold groups: they were younger and were more likely to be female, white collar/professional workers, and to have had higher education. They were also less likely to have had a previous transfusion or cardiac surgery.

Table 2 summarises the observed post-transfusion hepatitis events over the total time, and the results of the Mantel-Haenszel analysis comparing the no-withhold with the withhold group. This method of analysis combined the data from the two time periods (table 3) by averaging the two period specific odds ratios (one sided p=0.065 for overall post transfusion hepatitis). There was no evidence for heterogeneity of the surrogate testing effect between the pre-HCV and post-HCV screening periods at 5% significance, but there was a difference in the post-transfusion hepatitis rates between the two time-periods (Mantel-Haenszel test, p=0.011 for overall post-transfusion hepatitis and p=0.06 for HCV hepatitis, stratifying by centre and group).

There were no cases of HAV or HBV hepatitis in any of the study groups (95% CI, 0 to 1·3 per 1000 in either
Impact of anti-HCV screening on the benefit of NANB surrogate markers

We also analysed our data according to whether HCV screening was in place (table 3). Before HCV screening, the overall post-transfusion hepatitis rate per 1000 subjects was 20-2 in the no-withhold group compared with 5-0 in the withhold group (p=0.05). After mandatory HCV screening, the overall post-transfusion hepatitis rates were 8-6 and 6-8, respectively (p=0.55). The estimated benefit of NANB surrogate testing was 75% before HCV testing (95% CI, 1-7 to 15-6). No HCV hepatitis was seen in the syngeneic blood recipients (95% CI, 0 to 4-6 per 1000).

Discussion

During 1984–85, Feinman et al reported an overall incidence of NANB hepatitis of 92 per 1000 allogeneic blood product recipients. Subsequently, about one-third (31 per 1000) of these allogeneic blood recipients developed HCV hepatitis.1

During our study, withholding of NANB surrogate marker positive units reduced the overall post-transfusion hepatitis rate by 40%. Most of the benefit of NANB surrogate testing was due to a reduction in HCV hepatitis before blood was screened for HCV. During the period when screening for HCV was a requirement, the statistical power of our sample size was too low to detect moderate effects of NANB surrogate testing because of the extremely low event rate. The introduction of HCV screening thus appears to have modified the effect of NANB surrogate testing by reducing the risk of HCV hepatitis associated with exposure to NANB surrogate-marker-positive units. Nonetheless, our data suggest that NANB surrogate testing in Canada before May, 1990,
would have reduced the frequency of NANB hepatitis, especially that caused by HCV. By contrast, since HCV screening is now mandatory, the benefits of NANB surrogate marker testing are not supported by our data. Furthermore, the introduction of second and third generation HCV screening will probably decrease the HCV hepatitis risk further.

The drop in the HCV hepatitis rate from 31.3 per 1000 to 12.6 per 1000 between 1984–85 and 1988–90 appears to have been associated with improved methods for the screening of blood donors, since the drop occurred without NANB surrogate markers. In the USA a similar reduction in HCV hepatitis was reported over the same period in association with the introduction of NANB surrogate marker testing.15,16

Our study provides an interesting perspective on the potential nature of non ABC hepatitis. In at least 2 subjects cytomegalovirus was the causal agent. While it is possible that other non ABC hepatitis cases may have been caused by a hitherto unknown virus, from our study it appears that neither the introduction of anti-HCV screening nor the use of NANB surrogate markers had an impact on the frequency of non ABC hepatitis. Interestingly, 6-1 per 1000 recipients of syngeneic blood products had increases in alanine aminotransferase compatible with a diagnosis of this type of hepatitis. While hepatitis may be due to other viruses or to biologically active products present in stored blood (syngeneic or allogeneic), we believe that most cases with non ABC hepatitis are not transfusion related at all.

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References