VoxSanguinis
The International Journal of Transfusion Medicine
ISBT International Society of Blood Transfusion
Vox Sanguinis (2012) **102**, 82–90

INTERNATIONAL FORUM

DOI: 10.1111/j.1423-0410.2011.01506.x
Vox Sanguinis

International survey on NAT testing of blood donations: expanding implementation and yield from 1999 to 2009

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Introduction

Thirteen years ago, blood centres voluntarily started testing whole blood and platelet apheresis donors using nucleic acid amplification technology (NAT). The first reports on the feasibility of routine blood donation NAT screening were published in 1998/99, followed by wide-scale implementation or mandated NAT testing in multiple countries over the subsequent several years [1–5]. Although studies demonstrated that NAT testing could efficiently detect serologically negative donors who were infected with all three major transfusion-transmitted viruses (HCV, HIV-1 and HBV), most countries initially mandated NAT testing only for HCV or for HCV and HIV-1. Thus, despite the cost and logistical challenges to blood banks that tried to establish NAT testing on a routine basis, HCV and HIV-1 NAT testing expanded rapidly in the early 90s in many countries. The main reasons for the delayed implementation of HBV NAT were that the predicted yield and clinical value of interdiction of sero-negative HBV infections were low, commercial tests for routine NAT donor screening targeting all three viruses were not available at that time, and only a small number of blood banks had access to sensitive and reliable in-house HBV NAT tests.

An International Forum of Vox Sanguinis was organized in 2002 based on an eight-question survey entitled 'Implementation of donor screening for infectious agents transmitted by blood by nucleic acid technology'[6]. The experts in the field who responded to the survey did not necessarily represent countries where NAT testing had already been introduced. Questions referred to the viruses for which NAT testing was being performed or considered; the technology used; the pool size employed; the sensitivity, specificity and robustness of the assays; the degree of automation; the yield of NAT testing; the role of HCV core antigen testing; and the status of implementation of HAV and Parvovirus B19 NAT testing.

Because NAT screening was expanding and evolving very rapidly at that time, an updated survey was conducted in 2004 and published in 2005. This survey, which employed a standardized questionnaire based on similar questions to the 2002 survey, was sent to experts in 26 countries, with 18 countries contributed to this second International Forum on NAT testing [7].

Since the 2005 International Forum NAT screening has been introduced in many additional countries worldwide, hundreds of papers on sensitivity, specificity and yield of NAT testing have been published from many countries with diverse epidemiological situations resulting in dramatic differences in yield of window phase and occult infections. Assays and testing platforms have improved, with recent development of high-throughput automation enabling NAT testing of small pools or individual donations with reduced technical expertise and manpower. This important new technical approach to blood donation testing has significantly contributed to blood safety and provided new insights into the early dynamics of viral replication and infectivity of acute and chronic infections.

Methods

The Working Party on Transfusion Transmitted Infectious Diseases (WP-TTID) of the International Society of Blood Transfusion (ISBT) is dedicated to advancing blood safety in the world with specific focus on infectious risks. This

goal is accomplished through gathering and analysing relevant data and developing and coordinating international studies. In order to provide a convenient means to perform complex international surveys, the ISBT WP-TTID subgroup on virology developed an electronic questionnaire that can easily be distributed by e-mail and the responses readily compiled and analysed. It was decided to first use this tool to update the preceding International Fora on NAT testing. Although the survey included questions on overall yield since NAT testing was implemented, it was designed to focus on obtaining detailed cross-sectional data for one complete year of testing, rather than comprehensive longitudinal data over the whole testing period since the introduction of NAT in each individual country. This was because we wanted to compile and analyse data generated from current NAT technologies that are more sensitive and reliable than previous methods. These data, derived primarily using commercialized NAT systems, were expected to be more consistent such that results from different countries, including testing approaches, yield and epidemiology, could be compared.

The questionnaire contained detailed questions on the number of inhabitants and the donor populations in each country, the distributions of first-time and repeat donations, serological and NAT tests employed for screening, confirmation testing strategies, the viruses tested for by NAT and number of donations screened in 2008 and over the entire period since the date of introduction of NAT testing, whether NAT is mandated or not, the number of NAT-only-positive donations and the NAT yield rates, the NAT technology currently employed including pool size, sensitivity and specificity. Data were requested for whole blood and apheresis donations. Plasma for fractionation was excluded. The survey itself is available as supplemental material to this manuscript on the ISBT website (add link).

Results

The questionnaire was sent by e-mail in August 2009 to 77 experts from 59 countries. This distribution date seemed appropriate to allow each participant to compile and submit completed data including all confirmatory results for donations given in the 2008 calendar year. Most completed questionnaires were received between September and November 2009. A first electronic reminder was sent in November 2009, and a second personal reminder was sent to non-responding experts in May 2010. Seventy-three experts from 55 countries received the survey based on valid e-mail addresses, and 50 experts from 37 countries responded. Thirty-seven questionnaires from 25 countries were filled out completely and contained valid data for all questions including yield of NAT screening. In addition, 6 countries that did not perform NAT testing in 2008 sent back their serological data. Incomplete NAT data were received from seven other countries. The population covered by our survey (including the countries that did not perform NAT testing in 2008) totalled 1·2 billion. The population of all reporting countries in which NAT testing had been performed in 2008 totalled 1·16 billion. The exact numbers of donations with complete data sets for evaluation varied by virus as indicated in the respective sections that follow.

Implementation of NAT testing by country

Germany was the first country to introduce NAT screening of whole blood and apheresis donations on a routine basis with required negative NAT results prior to release of components (Fig. 1). Initially, this testing was voluntary and employed in-house NAT tests for HCV, HBV and HIV-1. HCV and HIV-1 NAT tests were mandated in Germany late in 1999 and 2004, respectively. Several other countries started NAT testing in 1999, primarily for HCV. Over the subsequent several years, NAT testing for HCV was mandated and began to be performed with commercial diagnostic assays that were more or less adapted to the needs of blood banks. Some countries started HIV-1 NAT testing as well. As seen in the Fig. 1, the number of countries per year that entered into HCV NAT testing was highest in 1999 and declined over time, whereas the number of countries that initiated HIV-1 NAT testing each year was relatively constant with small peaks in 2001–2003 and again in 2008. After the mandated introduction of HBV NAT in Japan and voluntary introduction of HBV NAT in Austria in 1999, there was a gap until 2004 followed by a peak in global implementation of HBV NAT in 2007–2008. In 2002, voluntary NAT for HBV was also introduced in several US blood centres (located in Sacramento California and Seattle Washington, with limited numbers of donations); broader implementation of HBV NAT did not occur in the United States until 2009 when multiplexed commercial NAT assays that included HBV detection were licensed in the United States.

As of 2010, 33 countries reported that they had introduced or would soon introduce HCV and HIV-1 NAT, and 27 of these reported current or planned introduction of HBV NAT. HCV NAT is mandated in 23 and HIV-1 NAT in 20 of the 33 countries assessed. HBV NAT is mandated in 12 and voluntarily performed in 15 countries.

NAT test systems and pool sizes employed in 2008

As technology improved significantly, early semi-automated and in-house-developed testing approaches have been generally replaced by fully automated commercial platforms and assays. Data on test systems employed were consequently requested for 2008 only (Table S1). The largest number of responding countries that have introduced NAT testing are in Europe. Thirteen of these

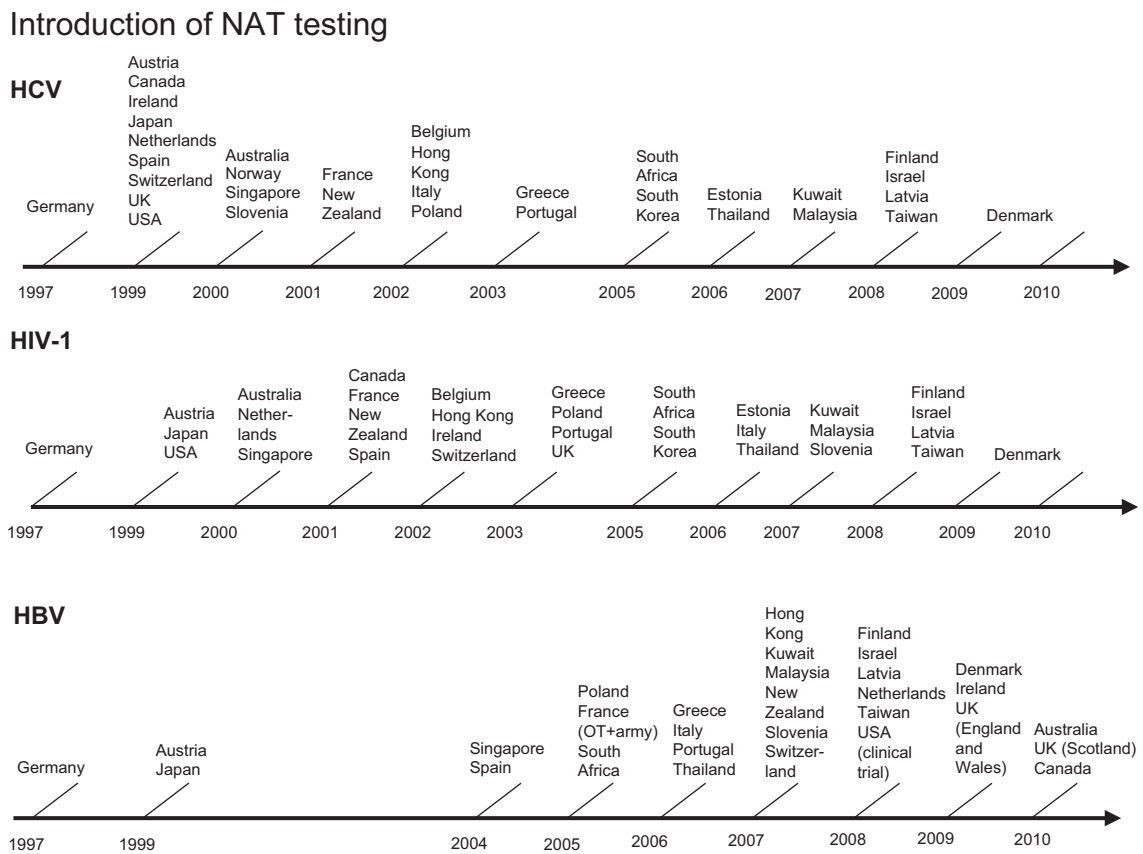

Fig. 1 Introduction of nucleic acid amplification technology (NAT) testing.

are using Roche assays and testing platforms, and 11 are using Novartis/Chiron assays/systems. Four countries [Germany, Austria, the Netherlands and UK (Scotland)] were using in-house testing approaches for the majority of their donations. Rest of UK were using Chiron. Germany, Austria and UK (Scotland) were the only countries in which in-house NAT tests were still in use in 2008 [UK (Scotland) subsequently converted to use of Roche's commercial assay]; the Netherlands performed commercial NAT assays with modified (in-house) extraction methods in 2008 but subsequently implemented the Roche extraction system.

In Africa, Asia-Pacific and North America, Novartis systems are the most prevalent (nine countries), followed by Roche (seven countries). None of these countries use in-house tests. Although there is no nationwide NAT testing in Brazil (it is planned for 2011 in the public system of hemocentres), one private hospital blood bank reported in-house NAT testing for HCV and HIV-1 since 1998, using an in-house system. It is estimated that ~5%–10% of donations in Brazil are now tested by NAT in private hospital blood banks for HIV-1, HCV and HBV using commercial or in-house adapted NAT assays.

In total, 22 countries perform Novartis/Gen-Probe NAT tests, 18 perform Roche NAT tests and only four perform in-house tests. In seven of the reporting countries, testing platforms from both commercial suppliers are approved and installed and operating in multiple testing laboratories. The numbers of donations tested by the respective testing systems do not reflect the distribution of numbers of tests applied per country due to variable penetrance of the different commercial NAT systems within countries. Some countries have blood centres that have introduced both commercial suppliers NAT assays/systems and centres that use in-house tests alone or in addition to commercial tests (Germany and Austria). Overall, a similar number of donations were tested by Roche (13 475 731) and Novartis (12 183 446) test systems in 2008, followed by in-house tests (3 258 028).

The majority of countries still performed NAT on mini-pools rather than individual donation testing in 2008, although there has been a clear progression towards smaller pools (e.g. 24 donations to six donations per minipool for Roche's system) and to individual donation testing (for Novartis/Gen-Probe's system). Discriminating numbers of donations tested and NAT yield data by pool size was not

feasible due to low numbers of donations tested in pools ≥ 48. Although there was a tendency to a lower yield of NAT positives among serologically confirmed positives for countries with high pool size especially for HCV and HBV, data were not conclusive. Germany with pools of 96 donations has reported comparable rates of virus detection (among both sero-negative and sero-positive donations) to those observed in epidemiologically similar countries performing HIV, HCV and HBV NAT on small pool sizes or even individual donations. This finding serves to emphasize the importance of sample processing and extraction techniques (e.g. ultracentrifugation of large volumes of pooled plasma as performed in Germany prior to extraction) in addition to pool size and analytical sensitivity of NAT assays on overall sensitivity and yield of NAT screening.

Yield of NAT testing in 2008

HIV-1

In total, 37·2 million donations were tested by HIV-1 NAT (Table S2). Complete data were received for 33·5 million donations (Table S3a). Of these 2189 donations (65·3/million) tested HIV-1 NAT positive, including RNA-positive donations with and without HIV-1 antibodies. The rate for first-time donations (295·5/million) was 11·2 times that for repeat donations (26·5/million). Almost two-thirds of the HIV-1 positives were reported from South Africa with 1331 cases (1796·7/million); the remaining 858 HIV-1 NAT positives were derived from the rest of the reporting countries with a composite rate of 26·2/million.

First-time donations from South Africa had a 22-fold higher HIV-1 NAT positivity rate (11 037·9/million) than repeat donations (503·2/million), whereas first-time donations (88·9/million) from the rest of the reporting countries had only a sixfold higher rate than repeat donations (15·4/million). It is anticipated that most of the HIV-1 NAT-positive repeat donors were acutely infected or incident infections, dependent on the interdonation intervals, whereas positive first-time donors reflect predominantly long-standing or prevalent infections. The rate (prevalence) of HIV-1-positive first-time donations in South Africa exceeded that from the rest of the reporting countries by a factor of 125, whereas the rate (incidence) of HIV-1-positive repeat donations in South Africa exceeded that from the rest of the reporting countries by a factor of 33. There are several countries in Asia and southern/eastern Europe with relatively high HIV-1 prevalence and incidence rates, reflected by overall NAT yield rates in first-time and repeat donors, respectively, compared with the majority of the remaining countries, but at far lower rates than for South Africa.

HIV-1 NAT-only positives. Compiled data from 2008 were reported for 37·4 million screened donations, of which 72 (1·9/million) were HIV-1 NAT-only positive (Tables 1 and S4a). Thirty-five of the 72 HIV-1 NAT-only positives reported in 2008 were from South Africa, with eight HIV-1 NAT-only positives identified in Spain and Thailand, five in the USA and four in Germany. All other countries reported no more than two HIV-1 NAT-only yield donations in 2008.

Data sorted by first-time and repeat donation status were provided for 35·3 million donations, with 4·9 million first-time and 30·4 million repeat donations. A total of 56 HIV-1 NAT-only positives were detected in these countries (overall rate of 1·6/million) with 19 HIV-1 NAT-only positive first-time donations (3·9/million) and 37 HIV-1 NAT-only positive repeat donations (1·2/million). Of these, 35 were reported from South Africa, with an HIV-1 NAT-only positive rate of 131·9/million for first-time donations and 35·4/million for repeat donations.

HIV-1 NAT positives among serologically confirmed positives. A total of 4917 HIV-1 serologically confirmed positive donations were reported out of 39·0 million donations tested (126·1/million) (Table S5a). Of these, 2174 sero-positive donations derived from 33·5 million screened (64·9/million) donations had corresponding HIV-1 NAT data assessable (Taiwan and Thailand reported serological data only) (Tables 2 and S6a). 2130 of 2174 (97·98%) sero-positive donations also tested HIV-1 NAT positive (Table 2). There was no major difference in the proportions of sero-positive donations with confirmed viraemia by NAT between first-time (97·84%) and repeat (98·24%) donations. Taking out the USA data that had lower rates of concordance of NAT with seropositivity (only 86·59% and 93·81% of HIV-1 sero-positive first-time and repeat donations tested NAT positive, respectively), the percentage of HIV-1 NAT positives among serologically confirmed positives increased to 99·29% and 98·91%, respectively, for the rest of the reporting countries. Very high rates of serologically positive donations were reported from Taiwan and Thailand without reporting NAT data.

The difference in the rates of HIV-1 serologically confirmed positive and HIV-1 NAT-only positives per million was a factor of 65·3. Taking out South Africa, a total of 3613 HIV-1 serologically confirmed positive donations were reported out of 38·2 million donations tested (94·5/million); restricting the analysis to these countries increased the ratio of serologically confirmed positive donations to HIV-1 NAT-only positives (1·01/million) to 93·6.

HCV

In total, 37·2 million donations were tested by HCV NAT (Table S2). Complete data were received for 26·6 million donations (no HCV NAT data were reported from Spain, and only combined data for first-time and repeat donations were reported from Japan) (Table S3b). Of these, 4586 (172·4/million) were HCV NAT positive, including RNA-positive donations with or without antibodies. The rate for first-time donations (995·2/million) was 35-fold that for

Table 1 NAT-only positives in 2008

Region/ country	Virus	First-time donations			Repeat donations			Total (first-time and repeat donations)		
		Number of tested donations	NAT-only positive	Rate/ 1 000 000 donations	Number of tested donations	NAT-only positive	Rate/ 1 000 000 donations	Number of tested donations	NAT-only positive	Rate/ 1 000 000 donations
Africa	HIV-1	90 959	12	131·93	649 864	23	35·39	740 823	35	47·24
	HCV	90 959	0	0·00	649 864	1	1·54	740 823	1	1·35
	HBV	90 959	11	120·93	649 864	21	32·31	740 823	32	43·20
Asia/Pacific	HIV-1[a]	1 362 593	1	0·73	8 195 982	2	0·24	10 053 686	11	1·09
	HCV[b]	811 646	9	11·09	3 369 691	4	1·19	9 753 686	19	1·95
	HBV[c]	185 979	10	53·82	542 805	12	22·11	5 805 840	101	17·40
Europe	HIV-1[a]	1 743 371	5	2·87	13 128 773	8	0·61	16 431 874	21	1·28
	HCV[b]	1 753 371	4	2·28	13 228 457	11	0·83	16 541 558	18	1·09
	HBV[c]	913 025	3	3·29	7 243 954	20	2·76	9 438 036	28	2·97
North America	HIV-1	1 678 862	1	0·60	8 408 812	4	0·48	10 087 674	5	0·50
	HCV	1 706 556	14	8·20	8 300 902	17	2·05	10 007 458	31	3·10
	HBV	702 533	1	1·42	3 200 417	7	2·19	3 902 950	8	2·05
South America	HIV-1	31 020	0	0·00	20 680	0	0·00	51 700	0	0·00
	HCV	31 020	0	0·00	20 680	0	0·00	51 700	0	0·00
	HBV	No NAT testing								
Total (all countries)	HIV-1[a]	4 906 805	19	3·87	30 404 111	37	1·22	37 365 757	72	1·93
	HCV[b]	4 362 532	27	6·19	25 569 594	33	1·29	37 095 225	69	1·86
	HBV[c]	1 892 314	25	13·21	11 637 040	60	5·16	19 887 649	169	8·50

NAT, nucleic acid amplification technology.

[a]Cumulative data only (first-time + repeat donations) were received from Thailand and Spain.

[b]Cumulative data only (first-time + repeat donations) were received from Kuwait, Thailand and Spain.

[c]Cumulative data only (first-.time + repeat donations) were received from Japan and Spain.

repeat donations (28·3/million). Estonia and Greece reported the highest prevalence in first-time donations with 7336·2/million and 4249·6/million, respectively, followed by Poland with 2886·3/million and Malaysia with 2858·3/million. Estonia and Greece also share the highest infection rates in repeat donations with Malaysia, 165·2, 134·2 and 193·3 per million, respectively; the HCV infection rate of donations from Polish repeat donors was about six-fold lower, with 23·9 HCV-RNA-positive donations/million. Taking out the data from Estonia, Greece, Malaysia and Poland resulted in a rate of 778·8/million HCV NAT-positive first-time donations and a rate of 25·2/million HCV NAT-positive repeat donations for the rest of the reporting countries. The difference between first-time and repeat donations decreased to a factor of 30·9 with the remaining countries, whereas for the indicated high-prevalence countries alone, the factor increases to a factor of 44·2.

HCV NAT-only positives. Compiled data were reported for 37·1 million donations in 2008, of which 69 (1·9/million) were HCV NAT-only positive (Tables 1 and S4b). Data sorted by first-time and repeat donation status were reported for a total of 29·9 million donations, including 4·4 million first-time and 25·6 million repeat donations. A total of 60 donations were HCV NAT-only positive (2·0/million). These HCV NAT yield donations sorted into 27 HCV NAT-only-positive first-time donations (6·2/million) and 33 HCV NAT-only-positive repeat donations (1·3/million). Due to low numbers of HCV NAT-only positives (see Tables 1 and S4b), further calculations were not made as they would not provide significant insights into the underlying epidemiology.

HCV NAT positives among serologically confirmed positives. A total of 13 903 HCV serologically confirmed positive donations were reported out of 38·6 million donations tested, for a rate of 360·3/million (Table S5b). Out of these, 6706 of 26·2 million (249·2/million) donations were assessable for comparison with NAT data and analysis of rates of nucleic acid detection among serologically confirmed positive donations by donation type (Taiwan and Thailand reported high numbers/rates of serologically positive donations without reporting HCV NAT data, and Japan reported only combined data for first-time and repeat donations). Of 6706 serologically confirmed positive donations, 4723 (70·4%) were also HCV NAT positive (Tables 2 and S6b).

Table 2 NAT positives among serologically positives in 2008

Region	Virus	First-time donations		Repeat donations		Total (first-time and repeat donations)	
		Number of tested donations	NAT positives among serologically positives	Number of tested donations	NAT positives among serologically positives	Number of tested donations	NAT positives among serologically positives
Africa	HIV-1	90 959	992/998 = 99·40%	649 864	304/306 = 99·35%	740 823	1296/1304 = 99·39%
	HCV	90 959	33/50 = 66·00%	649 864	16/29 = 55·17%	740 823	49/79 = 62·03%
	HBV	90 959	642/664 = 96·69%	649 864	68/68 = 100%	740 823	710/732 = 96·99%
Asia/Pacific	HIV-1	1 362 593	95/95 = 100%	8 195 982	138/138 = 100%	9 558 575	233/233 = 100%
	HCV [a]	734 314	425/566 = 75·09%	3 404 049	71/148 = 47·97%	4 138 363	496/714 = 69·47%
	HBV	110 352	709/818 = 86·67%	308 246	59/76 = 77·63%	418 598	768/894 = 85·91%
Europe – total	HIV-1	1 673 478	161/163 = 98·77%	11 698 433	181/185 = 97·84%	13 371 911	342/348 = 98·28%
	HCV	1 674 459	1523/2016 = 75·55%	10 390 293	125/180 = 69·44%	12 064 752	1648/2169 = 75·05%
	HBV	397 829	1132/1525 = 74·23%	4 259 078	172/293 = 58·70%	4 656 907	1304/1818 = 71·73%
Northern Europe	HIV-1	22 572	1/1 = 100%	255 425	1/1 = 100%	277 997	2/2 = 100%
	HCV	22 572	8/14 = 57·14%	255 425	2/3 = 66·67%	277 997	10/17 = 58·82%
	HBV	22 572	3/3 = 100%	255 425	2/2 = 100%	277 997	5/5 = 100%
Middle/Western Europe	HIV-1	1 065 791	40/42 = 95·24%	8 447 951	62/66 = 93·94%	9 513 742	102/108 = 94·44%
	HCV	1 065 791	306/455 = 67·25%	8 447 951	32/71 = 45·07%	9 513 742	338/526 = 64·26%
	HBV	266 153	376/442 = 85·07%	3 224 325	18/20 = 90·00%	3 490 478	394/462 = 85·28%
Eastern Europe	HIV-1	301 839	47/47 = 100%	1 491 574	39/39 = 100%	1 793 413	86/86 = 100%
	HCV	302 820	951/1202 = 79·12%	1 496 750	88/103 = 85·44%	1 799 570	1039/1305 = 79·62%
	HBV	72 242	688/1013 = 67·92%	589 161	151/270 = 55·93%	661 403	839/1283 = 65·39%
Southern Europe	HIV-1	283 276	73/73 = 100%	1 503 483	79/79 = 100%	1 786 759	152/152 = 100%
	HCV	283 276	258/345 = 74·78%	190 167	3/3 = 100%	473 443	261/348 = 75·00%
	HBV	36 862	65/67 = 97·01%	190 167	1/1 = 100%	227 029	66/68 = 97·06%
North America	HIV-1	1 652 996	144/166 = 86·75%	8 120 466	93/99 = 93·94%	9 773 462	237/265 = 89·43%
	HCV	1 627 850	2118/3049 = 69·47%	7 602 930	362/609 = 59·44%	9 230 780	2480/3658 = 67·80%
	HBV	1 250 280	145/217 = 66·82%	5 643 853	16/76 = 21·05%	6 894 133	161/293 = 54·95%
South America	HIV-1	31 020	13/14 = 92·86%	20 680	9/10 = 90·00%	51 700	22/24 = 91·67%
	HCV	31 020	30/35 = 85·71%	20 680	20/24 = 83·33%	51 700	50/59 = 84·75%
	HBV	No NAT testing					
Total (all countries)	HIV-1	4 811 046	1405/1436 = 97·84%	28 685 425	725/738 = 98·24%	33 496 471	2130/2174 = 97·98%
	HCV	4 158 602	4129/5716 = 72·74%	22 067 816	594/990 = 60·00%	26 226 418	4723/6706 = 70·43%
	HBV	1 849 420	2628/3224 = 81·51%	10 861 041	315/513 = 61·4%	12 710 461	2 943/3737 = 78·75%

NAT, nucleic acid amplification technology.

[a]HCV Asia/Pacific: data from Israel and Japan excluded (see Table S6b).

There was a great difference between rates of HCV RNA detection by NAT among sero-positive first-time (71·63%) and repeat (57·96%) donations. The ratio of serologically confirmed to NAT-only HCV positive donations was 193·7, which is approximately two times the respective factor for HIV-1.

HBV

In total, 20·9 million donations in 2008 were reported to have been tested by HBV NAT (Table S2). Complete HBV NAT data were received for 9·02 million donations (no HBV NAT data were reported from Spain, and only combined HBV NAT data from first-time and repeat donations were reported from Japan) (Table S3c). Of these, 3081 (341·70/million) were HBV NAT positive, including both serologically reactive and non-reactive donations. The rate for first-time donations (2151·4/million) was 42·6 times that for repeat donations (50·5/million). HBV NAT-positive donations from Greece and Malaysia accounted for 1517 of the 3081 cases, with a combined HBV DNA detection rate of 2169·8/million; when data from these countries were removed, there were 1564 HBV NAT positives among the remaining reporting countries, with a combined rate of 188·0/million.

First-time donations from Malaysia and Greece had 27·4-fold higher rates of HBV DNA detection (10 372·9/million) than repeat donations (378·2/million), whereas first-time donations (1 235·2/million) from the rest of the reporting countries had a 50·8-fold higher rate than repeat donations (24·3/million). The infection rates of Malaysian and Greek HBV-positive first-time donations exceed that from the rest of the reporting countries by a factor of 8·4, and the infection rates of HBV-positive repeat donations from these countries exceed that from the rest of the reporting countries by a factor of 15·6.

HBV NAT-only positives. Compiled data were reported for 19·9 million donations, of which 169 (8·5/million) were HBV NAT-only positive (Tables 1 and S4c). Split data for first-time and repeat donations were reported for a total of 13·5 million donations including 1·9 million first-time and 11·6 million repeat donations. A total of 85 donations were HBV NAT-only positive (HBV DNA-positive with negative HBsAg and anti-HBc, if performed), for a rate of 6·3/million. These sorted into 25 HBV NAT-only-positive first-time donations (13·2/million) and 60 HBV NAT-only-positive repeat donations (5·2/million).

Thirty-two HBV NAT yield donations were reported from South Africa, with a HBV NAT-only-positive rate of 120·9/million for first-time donations and 32·3/million for repeat donations, while 10 HBV NAT-only positives were identified in Hong Kong (47·0/million) and 9 (68·7/million) in Malaysia (Table S4c). It is important to recognize that these three countries, as well as many others, did not perform routine anti-HBc testing, and consequently, their yield numbers/rates include occult HBV infections (HBsAg negative, HBV NAT and anti-HBc positive), in addition to window period cases.

The above numbers do not include 79 HBV NAT yield donations (out of 5 077 238 combined first-time and repeat donations for a rate of 15·6/million) reported from Japan where anti-HBc screening is performed.

HBV NAT positives among HBsAg-confirmed positives. A total of 20 515 HBsAg-confirmed positive donations were reported out of 31·7 million donations tested for a rate of 647·3/million (no serological data were reported from Japan) (Table S5c). Out of these, 3737 HBsAg-positive donations detected following screening of 12·7 million donations (294·0/million) were assessable for comparison with HBV NAT data (Tables 2 and S6c). Of the 3737 HBsAg-positive donations, 2943 (78·75%) were HBV NAT positive. When sorted by donation status, 2628 of 3224 (81·51%) HBsAg-confirmed positive first-time donations and 315 of 513 (61·40%) HBsAg-confirmed positive repeat donations were HBV NAT positive (Tables 2 and S6c). Taking out the data from South Africa, Greece and Malaysia that had high HBV infection rates, HBV DNA was detected in 743 out of 899 (82·65%) HBsAg-confirmed positive first-time donations and 41 out of 106 (38·68%) HBsAg-confirmed positive repeat donations for the rest of the reporting countries.

The ratio of HBsAg-confirmed positives to HBV NAT-only positives was 121·4, similar to the NAT-only/serology yield ratio for HIV-1 and lower than the ratio for HCV. Taking out South Africa, Malaysia and Greece, a total of 16 021 HBsAg-confirmed positives were reported out of 28·7 million donations tested (557·7/million). This decreases the ratio of HBsAg-confirmed positives to HBV NAT-only-positive donations (6·0/million) to a factor of 92·95.

Anti-HBc screening was performed in 2008 by 12 of the 37 responding countries and partially by one more country (Table S7). However, data sets were often incomplete precluding rigorous evaluation. Focusing on the data on HBV NAT-only positives and HBV NAT and anti-HBc positives from those counties that reported data, either from anti-HBc screening or from anti-HBc confirmatory testing, there were a total of 75 HBV NAT-only-positive window period donations and 250 HBV NAT and anti-HBc-positive occult HBV infections detected (reported as HBsAg negative) out of 10 987 306 donations; this yields rates of 6·8/million for window period and 22·8/million for occult HBV infections, respectively. Although the yield numbers and ratios for individual countries were quite variable (Table S7), the data indicate that HBV NAT yield rates for occult HBV cases (HBV NAT and anti-HBc positive) generally exceeded the rates for acute window period infections (HBV NAT-only positives) by a factor of 3·35.

Yield of NAT testing since introduction

NAT yield data since introduction of NAT screening are summarized in Tables 3 and S8c. A total of 272 520 696 donations were screened by HIV-1 NAT, 303 196 074 were screened by HCV NAT, and 114 286 214 were screened by HBV NAT. Of these, 244 (0·9/million) were NAT-only positive for HIV-1, 680 (2·2/million) were NAT-only positive for HCV, and 1884 (16·5/million) were NAT-only positive for HBV DNA.

The number of screened donations and NAT yield data per country are also compiled in Table S8a. Compared with the NAT yield cases per number of donations tested in 2008 (Tables 1 and S4 a–c), there are no major differences in the total NAT yield rates since introduction of NAT testing for HIV-1, HCV and HBV, both regarding individual countries and geographic regions.

Additional findings

The survey requested that countries report the distribution of genotypes for HIV, HCV and HBV, both among the NAT yield donations and for the countries based on other epidemiological surveillance data. Although incomplete data

Table 3 NAT-only positives since introduction of NAT testing

Region/ country	Virus	Screened donations since implementation of NAT	NAT-only positives	NAT-only positives/ million
Africa	HIV-1	2 202 295	81	36·78
	HCV	2 202 295	4	1·82
	HBV	2 202 295	232	105·34
Asia/ Pacific	HIV-1	71 458 330	44	0·62
	HCV	71 458 330	169	2·37
	HBV	50 679 100	1091	21·53
Europe	HIV-1	110 860 111	73	0·66
	HCV	139 474 595	206	1·48
	HBV	56 342 555	550	9·76
North America	HIV-1	87 652 586	45	0·51
	HCV	89 652 687	299	3·34
	HBV	5 062 264	11	2·17
South America	HIV-1	347 374	1	2·88
	HCV	408 167	2	4·9
	HBV	No NAT testing		
Total (all countries)	HIV-1	272 520 696	244	0·9
	HCV	303 196 074	680	2·24
	HBV	114 286 214	1884	16·48

NAT, nucleic acid amplification technology.

were reported for a proportion of countries, the findings supported the conclusion that the genotypes present in blood donors are consistent with those reported from other studies in each country and region (Table S9).

In addition, the survey requested information on the HIV, HCV and HBV serological assays employed in donor screening, so the yield of NAT-only donations and the proportion of NAT positives among sero-positive donations could be evaluated in the context of the generation and manufacturer of respective serological screening tests (Table S10).

The survey asked participants to list the type and source of run controls used to validate and track performance of NAT assays (Table S11). Most countries performing NAT include such controls, generally manufactured by commercial vendors and supplied by the NAT vendors.

Finally, the survey requested information on the status of testing for other infectious agents, including HTLV-I/II, and NAT screening for Hepatitis A Virus, Parvo B19 Virus, West Nile Virus, Hepatitis E Virus and dengue virus (Table S12), as well as HBV vaccination programmes (Table S13).

Conclusions

This survey included 37 countries that reported results from NAT screening of approximately 300 million donations for HIV and HCV and over 100 million donations for HBV over the 10-year period extending from 1998 to 2008. To our knowledge, this is the largest data set ever compiled and most expansive study ever published on blood donor infectious disease screening.

The findings indicate remarkable progress in the implementation of molecular amplification techniques, with consequent interdiction of approximately 3000 viraemic donations that would have been missed by serological screening methods. The analysis demonstrated the successful expansion of NAT screening over the 10-year period, both in terms of numbers of countries screening and in terms of advances in technology and automation. As documented by the more detailed analysis of practices in 2008, the vast majority of NAT screening is now conducted with multiplexed HIV/HCV/HBV assay performed on highly automated instrument platforms that ensure reliable results with excellent sensitivity and specificity.

We believe that international bodies and regulators should more strongly encourage and preferably require NAT testing to improve blood safety (current recommendations and regulations from the World Health Organization, Council of Europe, Pan American Health Organization, etc., are not yet definitive in their recommendations for NAT testing). Although as our analysis confirms that the yield of NAT-only units is modest relative to the yield of serological screening, the infectivity of viraemic donations detected by NAT (with or without detectable serological markers) is very high. Hence, the relative impact of NAT screening is arguably greater than that of serological screening, although the existence of sero-positive but NAT-negative donations indicates that serological screening must be maintained even with the most sensitive NAT testing performed on individual donations. Consequently, the incremental cost-effectiveness of NAT is marginal since the safety benefits used in these calculations are restricted to the prevention of transmission of NAT-only yields and the cost of NAT testing remains relatively expensive.

The ISBT WP-TTID believes that collection and analysis of NAT and serological infectious disease donor screening data should be performed on a more regular basis in the future, given the expanded number of countries performing NAT, changing epidemiology of infectious diseases and donor selection practices and the likely evolution of NAT testing to target additional agents. Although this task is an appropriate one for the ISBT TTID working group, ongoing funding will be required to execute electronic surveys and rigorously analyse compiled data on a regular basis (every 3–5 years). The authors of this International Forum are grateful to the companies who have supported the working party to date and encourage their ongoing support, as well as funding from other sources (WHO, EU, NIH or CDC). Finally, we thank all of the

experts and the national blood transfusion services for their participation in completing the survey and reviewing the manuscript.

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Supporting information

Additional Supporting Information may be found in the online version of this article:
Data S1 Questionnaire on NAT testing of blood donations.
Table S1 NAT test systems and pool size applied in 2008
Table S2 Total number of donations tested by NAT in 2008
Table S3 (a) HIV-1 NAT, (b) HCV NAT and (c) HBV NAT positives in 2008
Table S4 (a) HIV-1 NAT-only, (b) HCV NAT-only and (c) HBV NAT-only positives in 2008
Table S5 (a) HIV-1, (b) HCV serologically positives and (c) HBsAg positives in 2008
Table S6 (a) HIV-1 NAT positives among serologically positives in 2008, (b) HCV NAT positives among serologically positives in 2008 including Israel and Japan and (c) HBV NAT positives among HBsAg positives in 2008
Table S7 Rates of HBV NAT-only positives and HBV NAT and anti-HBc positives, HBsAg negatives in 2008
Table S8 (a) HIV-1 NAT-only, (b) HCV NAT-only and (c) HBV NAT-only positives since introduction of NAT testing
Table S9 (a) HIV-1, (b) HCV and (c) HBV genotypes
Table S10 Serological test systems applied in 2008
Table S11 Run controls for HCV NAT
Table S12 Screening for other viral agents
Table S13 HBV vaccination programmes

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