

Development of the International Consortium for Blood Safety (ICBS) HCV panels

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إعداد رقائق مميزة لأنماط فيروس التهاب الكبد "سي" من قبل الاتحاد الدولي لسلامة الدم
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الخلاصة: قام الاتحاد الدولي لسلامة الدم ICBS بإعداد رقائق Panels تمييزية لأنماط فيروس التهاب الكبد "سي" لتقييم ما تتمتع به المقاييس المستخدمة لتحري أضداد فيروس التهاب الكبد "سي" من حساسية ونوعية. وقد قام الاتحاد بجمع وتوصيف 1007 وحدة من البلازما الإيجابية لفيروس التهاب الكبد "سي" من مناطق تختلف اختلافاً جغرافياً واسعاً، مستخدماً مقياساً المعتمد المناعي المرتبط بالإنزيم ELISA ومقاييس التخطاط المناعي المشوب RIBA والتفاعل السلسلي للبوليميراز لإنزيم المنتسخة العكسية RT-PCR وتعيين النمط الجيني استناداً إلى المتواليات، وقد وجد أن 539 وحدة منها كانت ملية لمعايير تحديد الإيجابية الحقيقية، وتم انتقاء مئتين من بين هذه الوحدات المؤكدة الإيجابية تمثل الأنماط الجينية الستة الرئيسية لتجميع المكونات الإيجابية الحقيقية لكل رعل. أما الرقائق السلبية فتتألف من 181 وحدة بلاسمية جمعت من الولايات المتحدة الأمريكية. وقد أثبتت الرقائق أنها تتمتع بقيمة كبيرة في تعيين أداء مقاييس أضداد فيروس التهاب الكبد "سي"، مما يمكن السلطات الوطنية، ولاسيماً في البلدان المحدودة الموارد من اتخاذ قرارات مستنيرة حول انتقاء مقاييس يمكن الوثوق بها ويسهل دفع تكاليفها.

ABSTRACT To evaluate the sensitivity and specificity of assays used to screen blood for antibody to hepatitis C virus (HCV) infection, the International Consortium for Blood Safety (ICBS) established fully characterized ICBS panels. ICBS collected and characterized 1007 anti-HCV-positive plasma units from geographically diverse origins by ELISA, RIBA, RT-PCR, and sequence-based genotyping, 539 of which met the definition of a true positive. Of these, 200 confirmed positive plasma units, representing the 6 major HCV genotypes, were selected to assemble the true-positive constituents of the panel. The negative panel comprises 181 plasma units collected from the USA. The panels have proved valuable for determining the performance of anti-HCV assays thus permitting national authorities, especially in resource-limited countries, to make informed decisions on selection of affordable and reliable assays.

Création par l'International Consortium for Blood Safety (ICBS) de panels pour le dépistage du VHC
RÉSUMÉ Afin d'évaluer la sensibilité et la spécificité des tests utilisés pour effectuer une recherche d'anticorps dirigés contre le VHC (virus de l'hépatite C) dans le sang, l'International Consortium for Blood Safety (ICBS) a créé des panels correctement définis. L'ICBS a recueilli et caractérisé 1007 unités de plasma anti-VHC positives provenant de zones géographiques diverses en utilisant les tests ELISA, RIBA, RT-PCR et le génotypage par l'analyse des séquences ; 539 d'entre elles correspondaient à la définition d'un vrai positif. Sur ces unités, 200 confirmées positives représentant les six principaux génotypes VHC ont été sélectionnées pour constituer les vrais positifs du panel. Le panel négatif est constitué de 181 unités de plasma recueillies aux États-Unis. Les panels se sont avérés utiles pour déterminer la performance des tests anti-VHC, ce qui a permis aux autorités nationales, notamment dans les pays disposant de ressources limitées, de choisir en connaissance de cause des tests abordables et fiables.

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Introduction

Infection with hepatitis C virus (HCV) is a global health problem, with an estimated 170 million chronically infected persons worldwide. Prevalence varies greatly by region and country. Countries in Africa and Asia have the highest reported HCV infection prevalence rates, whereas Japan, Australia, and the industrialized countries of North America and Europe have relatively low prevalence. Rates range from $\geq 22\%$ in Egypt, the country with the highest reported prevalence, to as low as 0.6% in Germany. Persons with persistent HCV infection are at high risk for development of chronic liver disease, cirrhosis and hepatocellular carcinoma [1,2].

Sources of HCV infection include: unsafe injections, transfusion of blood or blood products from unscreened or inadequately screened blood donors, transfusion of blood products that have not undergone viral inactivation, long-term haemodialysis, organ transplantation, and medical or dental procedures performed with inadequately sterilized instruments. An unsafe blood supply represents a major contributor to the total HCV disease burden in many developing countries. The World Health Organization (WHO) estimates that blood donations up to 13 million units of the global blood supply, mainly in low or medium Human Development Index countries, are not screened for all relevant transfusion-transmissible infections [3].

Three main factors determine the efficiency of anti-HCV diagnostics: 1) target antigens used in the assay design, 2) professional skill of the technician performing the assay, and 3) laboratory environment. In many countries, test kits must be licensed by a national control authority before they can be used by blood transfusion services to screen donated blood. Licensure might

involve testing each kit against a panel of specimens obtained from the general population to determine the kit's sensitivity and specificity compared to a reference test. In many developing countries, test kits are licensed based on their licensure elsewhere in the world. Furthermore, because HCV can be classified into 6 major genotypes and more than 60 subtypes, test kits should be assessed for their accuracy and consistency across the range of known HCV genotypes and subtypes.

In resource-challenged countries, the expense of currently available assays for blood screening results in a lack of or inconsistent testing of blood donations. In addition, transfusion services and laboratories are hampered by the generally poor specificity of anti-HCV screening assays. These 2 constraints underscore the need to identify diagnostic test kits that are sensitive and specific and also affordable.

The International Consortium for Blood Safety (ICBS), a charitable non-for-profit organization operational since 1998, has specific goals, which include the identification and evaluation of reliable and affordable assays for screening of blood donations. In line with its goals, ICBS decided to establish HCV panels (as well as the ICBS HBV panels) for comparative evaluation of commercially available test kits. The ICBS HCV panels were developed at the ICBS Collaborating Center at the Division of Viral Hepatitis, Centers for Disease Control and Prevention (CDC), Atlanta, United States of America (USA), with the assistance of the former Visible Genetics Inc., Norcross, Georgia, USA. The aim of creating the HCV panels was to provide fully characterized panels of anti-HCV-positive and -negative samples that can be used to determine the sensitivity and specificity of test kits manufactured anywhere in the world.

Methods

Plasma specimens

A total of 1007 anti-HCV initially reactive (IR) plasma units were obtained from collaborators in Brazil, Egypt, Indonesia, Ivory Coast, South Africa, the USA and Viet Nam. Certain regions were selected based on the prevalence of unique genotypes and subtypes to ensure their inclusion in the ICBS Panel.

Each collaborator was at the start asked to store anti-HCV IR specimens. After permission to export from each participating country was obtained, all IR plasma units were shipped frozen, on dry ice, to the ICBS Collaborating Center (Division of Viral Hepatitis, CDC) where they were characterized. All of the samples received were unlinked; no information related to the blood donors was provided. Units were selected for inclusion in the ICBS Panels based on: 1) the definition of true positivity, 2) the diversity by genotype and subtype, 3) the confirmatory assay banding patterns (RIBA 3.0, Chiron, Emeryville, California, USA), 4) the geographic region, and 5) the volume of material. The original strategy was to prepare anti-HCV ICBS Panels composed of approximately an equal number of panel constituents across all genotypes.

The ICBS collaborators from developing countries faced obstacles and failed in getting permission from the authorities in their countries to export normal negative plasma units to ICBS for the inclusion in the ICBS negative panel. Therefore, for specificity determination, a total of 181 plasma units negative for markers of HCV (anti-HCV), hepatitis B virus (HBV; HBsAg and anti-HBc), human immunodeficiency virus (HIV; anti-HIV-1/2 and HIV-1 p24 Ag), human T-cell lymphotropic virus (HTLV; anti-HTLV I/II), syphilis, and parvovirus B19 (DNA PCR) infection were acquired

from the USA. These plasma units were also screened for HCV and HIV by nucleic acid tests using Procleix HIV-1 and HCV/nucleic acid pooled testing (Gen-Probe Inc., San Diego, California, USA). In addition, ICBS constructed 19 pooled units (each assembled by pooling 2 to 4 small-volumes plasma units from the same country) from IR samples when tested in their countries of origin. These IR samples were obtained from Indonesia, Egypt and Ivory Coast that were subsequently found to be negative by retesting at CDC. However, additional testing using these 19 pooled samples demonstrated an unacceptable number of false-positive results with many of the test kits evaluated and therefore these pooled samples were omitted from the final panel.

Characterization of plasma units

All IR units were retested at CDC by an anti-HCV screening assay (Ortho EIA 3.0, Ortho-Clinical Diagnostics, Raritan, New Jersey, USA) and positive specimens were retested in duplicate. Repeatedly positive units were confirmed by automated RIBA 3.0 and by quantitative reverse-transcription polymerase chain reaction (RT-PCR) (Roche Amplicor, Roche Molecular Systems, Roche Diagnostic Systems Inc., Branchburg, New Jersey, USA).

Viral RNA isolation

HCV-specific RNA was extracted from 100 μ L of serum or plasma using Tripure isolation reagent (Roche Applied Science, Indianapolis, Indiana, USA), according to the manufacturer's protocol.

Reverse transcription (RT)

A 17- μ L per sample RT mix, containing 2 μ L of 10X PCR buffer without magnesium chloride (Perkin-Elmer), 4 μ L of 25 mM magnesium chloride, 8 μ L of 2.5 mM deoxynucleotide-triphosphate (dNTP),

and 2 µL of random primers and 1 µL of reverse primer, was prepared and mixed with the air-dried RNA pellet. The sample was heated at 94 °C for 3 minutes, rapidly frozen on dry ice, thawed on ice, and then centrifuged at 2000 rpm for 1 minute; 3 µL of the RT enzyme mix (1.5 µL of RNase- and DNase-free water, 1 µL of AMV-reverse transcriptase, and 0.5 µL of RNase inhibitor) was added to the sample. After incubation at 42 °C for 60 minutes and heating at 95 °C for 5 minutes, the sample was chilled on ice.

Nested RT-PCR and sequencing

Nested RT-PCR was performed according to the method described by Nainan, Cromeans and Margolis [4].

Primer sequences

Primer sequences for the 5'UTR region were synthesized as follows: outer-forward, 5'GTG CAC GGT CTA CGA GAC CT; outer-reverse, 5'CTG TGA GGA ACT ACT GTC T; inner-forward, 5'CCC TAT CAG GCA GTA CCA CAA; inner-reverse, 5'GAA AGC GTC TAG CCA TGG CGT.

Primer sequences for the NS5B region were as follows: outer-forward, 5'TGG GGA TCC CGT ATG ATA CCC GCT GCT TTG A; outer-reverse, 5'GGC GGA ATT CCT GGT CAT AGC CTC CGT GAA; inner-forward, 5'CTC AAC CGT CAC TGA GAG AGA CAT; inner-reverse, 5'GCT CTC AGG CTC GCC GCG TCC TC.

Genotype analysis by nucleotide sequencing and phylogenetic analysis

All confirmed anti-HCV-positive units were genotyped and subtyped by direct DNA sequencing of the NS5b region and the 5'-UTR region, both by CDC and by

the former Visible Genetics Inc. Genotype/subtype assignment was based on the classification of Simmonds et al. [5]. Additional genotype/subtype assignment was conducted on some of anti-HCV units based on the methods of Tokita et al. [6] (full data available at <http://www.emro.who.int/publications/emhj/index.asp>). The sequences obtained in this study were analysed by both homology- and phylogenetic-based methods. Sequences were edited using BioEdit and aligned with ClustalX. Genotyping (including subtyping) was done using the neighbour-joining method in Clustal X with a set of reference sequences from each genotype that clustered with the test sequences to form specific clades. All the 200 constituents of the HCV panel were genotyped by CDC and many of these constituents (126 out of 200) were also genotyped by the former Visible Genetics Inc. Thus in total 126 panel constituents were assigned a genotype and subtype based on concordant results from the 2 independent laboratories (CDC and former Visible Genetics Inc.).

Establishment of the HCV ICBS

Panel specimens

It was estimated by a special ICBS task force for establishing the ICBS panels that, for the purpose of evaluating anti-HCV assays, an HCV panel of 200 genotyped constituents is reasonably suitable for comparative evaluation of the sensitivity of commercially available anti-HCV assays. The 200 constituents of the ICBS anti-HCV Panel were selected from 539 specimens that met the definition of a true positive. A true-positive specimen was defined as one that was repeatedly reactive by the screening assay, RIBA 3.0 positive, and positive in the RT-PCR-based genotyping assay; specimens classified as indeterminate by RIBA 3.0 did not meet the definition. Specimens were selected for the ICBS Panel based on: 1) serologic

confirmation as positive (indeterminate specimens were excluded); 2) concordant genotype/subtype if possible (i.e. genotyping conducted by both CDC and the former VGI); 3) RIBA 3.0 banding patterns; 4) geographic origin; and 5) volume of ≥ 100 mL. Plasma units meeting the selection criteria were manufactured into panels under contract according to Good Manufacturing Practice/Good Laboratory Practice.

The required minimum volume of each plasma unit to manufacture the HCV panel samples was 200 mL. However, the volume of most plasma units obtained from certain geographic areas, such as Indonesia and Viet Nam, was < 200 mL because of the small volume (approximately 250 mL) of blood units collected by blood banks in these countries. Plasma units with volumes < 200 mL selected for the panel were brought up to the required 200-mL volume by adding plasma that was negative for HIV, HBV and HCV markers. The addition of the negative plasma to bring up the volume of some of the anti-HCV panel samples to the required 200-mL volume did not affect their detection by anti-HCV assays. All manufactured units in the anti-HCV panel and the negative panel were retested and confirmed to meet the criteria of "true positive" and "true negative" respectively.

Inclusion of weak-positive samples in the ICBS HCV panel

The ICBS HCV panel includes weak-positive samples (optical density reading close to the cut-off point of the assay) with signal to cut-off (s/co) ratios low enough to challenge the sensitivity of the anti-HCV assays being evaluated and distinguish between sensitive and less sensitive assays. Post-manufacturing use of the 200 ICBS HCV panel members to evaluate the sensitivity of 44 commercial ELISA as-

says, showed that 19 HCV panel specimens (panel members number: 169, 33, 76, 156, 166, 12, 178, 67, 18, 198, 54, 121, 172, 73, 116, 164, 79, 104 and 13 in the order of the number of false negatives with the 19 HCV samples) are weak positives having screening-test-positive signals close to the cut-off where differences in the antibody reactivity would be more likely to be seen. These 19 anti-HCV panel samples gave false negative results in ≥ 2 ELISA assays of the 44 test kits evaluated (full data available at <http://www.emro.who.int/publications/emhj/index.asp>).

Inclusion of commercial seroconversion panels

The ICBS anti-HCV Panel was supplemented with 3 commercially available HCV seroconversion panels for use for the ICBS comparative performance studies of anti-HCV assays [7]. These 3 seroconversion panels are: PHV 914, PHV 920 (BBI Diagnostics, West Bridgewater, Massachusetts, USA) and HCV 6212 (Impath-BCP, Franklin, Maryland, USA). The panels were selected to represent different reaction patterns, i.e. core only (PHV914), NS3 only (HCV 6212) and core + NS3 (PHV920) and to include different genotypes, i.e. genotype 1a (PHV 920) and genotype 2b (PHV 914). Characterization data were taken from the data sheets of the respective panel suppliers but are all based on Chiron RIBA HCV 3.0 Strip Immunoassay (Chiron Corporation, Emeryville, California, USA).

Storage of ICBS panels

The ICBS panels are stored at -70°C in the ICBS Test-Kit Evaluation Center, at Paul-Ehrlich-Institut (PEI), Langen, Germany, where the test kits evaluations were performed.

Results

True-negative panel

The anti-HCV-negative panel was assembled in June 2002. As mentioned earlier in this paper, the ICBS collaborators faced difficulties which prevented shipping normal negative plasma from their countries to ICBS. The 19 samples constructed by pooling 2–3 small units (IR in countries of origin and subsequently found to be negative) obtained from Indonesia, Egypt and Ivory Coast were excluded from the final negative panel. They were disqualified based on the high rate of false-positivity in many anti-HCV assays and the requirement for use of verified, unequivocally “true” and non-pooled negative samples in ICBS panels. Therefore, all of the 181 unequivocal anti-HCV-negative samples included for evaluation of the specificity of anti-HCV test kits were obtained from the USA. All constituents of the true-negative panel were negative for HCV, HBV, HIV, HTLV, syphilis and parvovirus B19.

True-positive panel

Of the 1007 plasma units received from collaborating transfusion centres, only

539 (54%) met the definition for a true-positive sample (Table 1). The most common genotype in the 539 samples was genotype 1 ($n = 296$), followed by genotype 4 ($n = 129$), genotype 6 ($n = 43$), genotype 3 ($n = 42$), genotype 2 ($n = 26$), and genotype 5 ($n = 3$).

Although the original strategy was to produce an anti-HCV panel composed of 200 components with approximately equal representation of all genotypes, this goal was not achieved due to difficulties in obtaining sufficient numbers of specimens belonged to genotypes 2 and 5 that met the selection criteria. The characteristics of the 200 constituents of the ICBS anti-HCV panel can be seen on line at <http://www.emro.who.int/publications/emhj/index.asp>. The post-manufacturing quantitative PCR results for 22 of the 200 constituents of the HCV-positive panel (11 from Viet Nam, 8 from Indonesia, 2 from Egypt and 1 from Brazil) have < 600 IU/mL, due to the dilution effect after bringing the constituents to the required 200-mL volume by adding negative plasma. These 22 constituents, however, tested positive in the RT-PCR assay used for genotyping.

Table 1 Distribution of hepatitis C virus (HCV) genotype by country among the true anti-HCV positive plasma units collected

Country	HCV genotype						Total
	1	2	3	4	5	6	
Brazil	46	1	12	0	0	0	59
Egypt	6	0	1	124	0	1	132
Indonesia	91	13	21	2	0	0	127
Ivory Coast	1	0	0	0	0	0	1
South Africa	0	0	0	0	3	0	3
United States of America	45	8	4	2	0	0	59
Viet Nam	107	4	4	1	0	42	158
Total	296	26	42	129	3	43	539

Table 2 shows the number of ICBS anti-HCV panel constituents by genotype and country of origin. The most diverse genotype by country of origin was genotype 1, with 66 constituents representing 6 countries from different regions of the world. Genotype 2 constituents originated from 4 countries, but only 23 samples met the criteria for inclusion in the anti-HCV panel. Genotype 3 constituents also originated from 4 countries; 40 of the true-positive units were included. The 36 constituents belonging to genotype 4 were obtained from 4 countries, but most originated in Egypt. All except one of the genotype 6 constituents were obtained from Viet Nam, and all 3 constituents belonging to genotype 5 were from South Africa. The constituents of the anti-HCV panel for genotypes 4, 5, and 6 did not reflect such diversity because they were obtained from regions where the respective genotype is most prevalent compared to other regions.

Discussion

The genotyped ICBS anti-HCV Panel

An ICBS anti-HCV Panel was established as a “real world” panel to determine the

performance of individual test kits but not to monitor lot quality or intra-assay variability on a regular basis. The ICBS Panel was assembled to facilitate the evaluation and identification of reliable and affordable HCV test kits.

The ICBS HCV panel, composed of 200 unequivocally confirmed anti-HCV positive constituents collected from diverse geographical regions of the world selected on the basis of the prevalence of genotypes and subtypes to ensure their inclusion in the panel, provides details on genotypes, s/co values of the screening assay (Ortho HCV 3.0), antibody profile (HCV RIBA 3.0) and PCR result (quantified). Yet the variability in characteristics and sequences of the HCV panel constituents may have been restricted by the sensitivity of initial ELISAs and by the sequences detected by the molecular assays used for genotyping, which may cause a probable exclusion of some variants. However, the ICBS HCV panel potentially serves as an alternative (or complementary panel) to other panels used to perform comparative performance studies of anti-HCV assays. This panel was used together with commercial seroconversion panels in the evaluation of 44 assays for

Table 2 Distribution of hepatitis C virus (HCV) genotypes among the selected HCV panel constituents

Country	HCV genotype						Total
	1	2	3	4	5	6	
Brazil	13	1	0	0	0	12	26
Egypt	4	0	0	31	0	1	36
Indonesia	16	11	21	2	0	0	50
Ivory Coast	1	0	0	0	0	0	1
South Africa	0	0	0	0	3	0	3
United States of America	7	8	4	2	0	0	21
Vietnam	25	3	3	1	0	31	63
Total	66	23	28	36	3	44	200

antibodies to HCV in addition to in addition to 5 other assays licensed in the European Union (EU), used for comparison. The true-positive ICBS Panel has already proven valuable for the selection of high-sensitivity screening assays manufactured in the developing world and countries in transition and the substantial variability in sensitivity and specificity among the assays documented the value of the panel [7]. Continued use of the ICBS Panel to evaluate the performance of anti-HCV assays will fill an important need by identifying reliable and affordable test kits that can be used in countries with limited resources.

Evaluations of test kits to date have shown no obvious genotype-specific detection failures, indicating that current assays do not appear to be genotype specific [7]. Thus, the lack of equal representation of all genotypes on the ICBS Panel might not affect sensitivity determinations.

Comparison to other HCV panels used for comparative evaluation of anti-HCV assays

Other HCV panels used by other institutions to perform evaluation studies of anti-HCV assays include the panels used by WHO and the National Institute of Infectious Diseases (NIID), Toyama, Shinjuku, Tokyo, Japan [8–10]. The WHO panel includes 68 serologically anti-HCV positive specimens (1 from Africa, 20 from Asia, 40 from Europe and 7 from Latin America). The specimens in the WHO panel were screened by 2 reference ELISAs (Ortho 3.0 Enhanced SAve from Ortho Clinical Diagnostics and Monolisa anti-HCV Plus from Bio-Rad). Specimens showing reactivity with either or both ELISAs were further characterized by using Chiron HCV RIBA 3.0. When results of the reference ELISAs and the HCV RIBA 3.0 were all positive, a specimen was considered anti-HCV positive. Similarly, speci-

mens showing discordant ELISA reactivity and a positive HCV RIBA 3.0 were considered anti-HCV positive. When the initial ELISA results were discordant, specimens having negative HCV RIBA 3.0 results were considered anti-HCV negative. As it is the case with the ICBS panel specimens, WHO specimens with discordant ELISA results and indeterminate HCV RIBA 3.0 were excluded from the panel. The WHO panel was used to evaluate 5 simple/rapid anti-HCV tests (report 1) followed by the evaluation of an additional 5 anti-HCV assays (2 simple/rapid tests and 3 ELISAs) with the inclusion of 4 reference assays for comparative purposes (report 2) [8,9].

As for the HCV panel used by NIID, Japan, to re-evaluate 25 anti-HCV test kits approved for marketing in Japan, it is composed of 20 NIID HCV antibody positive sera from the Institute, 1 commercial low titre panel and 2 commercial HCV antibody seroconversion panels.

Limitations of the negative panels used for comparative evaluation

The ICBS HCV negative panel used to investigate commercial anti-HCV assays is composed of 181 samples, while the WHO's anti-HCV negative panel and that of NIID, Japan, are composed of 189 and 19 samples respectively. These panels can only provide a limited specificity estimate. Still, the ICBS HCV negative panel gave an impression on the general specificity that could be expected by the assays [7]. Some centres, contemplating the introduction of a test, evaluate specificity by performing ≥ 2000 tests. Specificity found in 1 country may not necessarily be similar in another. Since different countries may have their own "false-positive populations", specificity is best determined locally by inclusion of some negative samples from the local population. The false-positive results of the

19 pooled samples might have been the result of pooling, hypergammaglobulinaemia, connective tissue disorders, or other factors related to the origin of these samples. The questionable pooled samples were derived from countries in which malaria and other parasitic infestations are common. It was presumed in other studies that stimulation of polyclonal antibody production from recurrent malaria infection could produce false-positive anti-HCV enzyme immunoassay results, thereby generating a high false-positive rate [11–13]. Nonetheless, when used in the tropics, the negative panel might underestimate the expected number of false-positive results. Supplementing future panels with additional samples from tropical regions will assist in alleviating this problem.

Need for affordable confirmatory assays

In some resource-limited countries, a high rate of false-positive findings might limit the blood supply available for transfusions, leading to increased adverse patient outcomes. Because the use of the RIBA assay as a confirmatory test might not always be possible due to its cost, affordable confirmatory assays should be developed and made available for countries with limited resources. Identification and establishment of regional confirmatory laboratories capable of performing RIBA assays, nucleic acid testing by RT-PCR, and/or testing for HCV core antigen by ELISA would also be highly advantageous. HCV core antigen can be detected during the window phase of infection and appears to be a suitable additional marker for screening of blood donations [14–16].

Conditioned availability of ICBS HCV panel samples

In line with the strong belief of ICBS in capacity-building, developing countries

that have national control authority laboratories could initially be offered, at no cost, small samples of the ICBS Panel to help them develop their panels for continued use. These control authority laboratories should have appropriate facilities and resources, including properly trained personnel able to validate candidate tests and committed to develop their own country/region specific panels (in phases) according to a well-defined plan and time frame. In spite of the importance of including seroconversion panels in national HCV panels to evaluate diagnostic sensitivity in assays and estimate delay in detection of anti-HCV, seroconversion panels are very expensive and their high cost may hinder their inclusion in national panels of many countries with restricted economies.

Use of the ICBS HCV Panel by the ICBS Test Kits Evaluation Center

The great majority of countries in the developing world do not have regulatory institutions capable of properly evaluating test kits. Therefore, ICBS recognized the necessity of establishing a centralized test-kit evaluation centre in an attempt to fill the gap. ICBS and Paul-Ehrlich-Institut in Langen, Germany agreed to collaborate in this regard. The Paul-Ehrlich-Institut agreed to foster the “ICBS Blood Screening & Test-Kits Evaluation Centre” within its framework. This centre is carefully evaluating available commercial assays using the ICBS panels. Information on sensitivity and specificity of evaluated assays are posted on the ICBS website (www.icbs-web.org). This information, together with other available information, may help countries with no control authority laboratories in making informed decision for selecting reliable and affordable assays to be used by blood banks for screening of blood donations.

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AIDE-MEMOIRE for national blood programmes

A well-organized blood transfusion service, with quality systems in all areas, is a prerequisite for the safe and effective use of blood and blood products.

The HIV/AIDS pandemic has focused particular attention on the importance of preventing transfusion-transmitted infections. Recipients of blood products are also infected by hepatitis B and C viruses, syphilis and other infectious agents, such as Chagas disease. The global burden of disease due to unsafe blood transfusion can be eliminated or substantially reduced through an integrated strategy for blood safety, which includes:

establishment of a nationally-coordinated transfusion service

- collection of blood only from voluntary non-remunerated blood donors from low-risk populations
- testing of all donated blood, including screening for transfusion-transmissible infections, blood grouping and compatibility testing
- reduction in unnecessary transfusions through effective clinical use, including the use of simple alternatives, wherever possible.

This aide-mémoire is available in English, French, Portuguese and Spanish, and can be accessed from: <http://www.who.int/bloodsafety/en/>.