Screening Donated Blood for Transfusion Transmissible Infections in Bangladesh

National Guidelines 2013
Mandatory screening of donated blood for transfusion in Bangladesh began in 2000 after implementation of the Safe Blood Transfusion Programme (SBTP) at all hospital based blood transfusion centres. Testing began under the purview of the Safe Blood Transfusion Act 2002 which states that before transfusion, all blood and blood products must undergo testing for five transfusion transmissible infections (TTIs), i.e. Hepatitis B, Hepatitis C, syphilis, malaria and HIV. However, nationally agreed strategies for screening were not in place, nor were policies on how results of screening should be handled or how donors should be managed regarding the outcome of testing. Strategies on the types of assays in the context of facilities, infrastructure and manpower, as well as throughput of blood collection at individual centres, were not available. Centres able to provide confirmatory testing, and to which blood samples for further confirmation of initial reactive results could be referred for donor notification, counselling and care, were not defined. For an effective donor blood screening programme, national screening strategies should be developed to give strategic direction based on country context and need.

In addition, strategies should be regularly reviewed to take cognisance of new assays, changes in manpower, particularly technologists, and changes in infection marker epidemiology. The blood screening strategy must also define a testing algorithm to provide uniformity in the standard of testing and management of reactive cases for counselling. The aims are to establish a reliable blood screening programme and management of reactive donors and to prevent the transmission of infectious diseases by transfusion.

In the preparation of this document (protocol), the context, principles and strategies were discussed with experts on the national blood transfusion expert committees of the Ministry of Health. This protocol may be incorporated into the national blood screening programme for the establishment of referral centres for validation of kits, preparation of annual procurement plans for kits, modality of transportation of kits to centres, and validation of algorithms applicable to TTI reactive cases.
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## Acronyms

<table>
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<th>Acronym</th>
<th>Description</th>
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<tr>
<td>BTS</td>
<td>Blood Transfusion Service</td>
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<tr>
<td>CLIA</td>
<td>Chemiluminescent Immunoassay</td>
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<td>EIA</td>
<td>Enzyme Immunoassay</td>
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<td>EQAS</td>
<td>External Quality Assessment Scheme</td>
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<td>HBsAg</td>
<td>Hepatitis B Surface Antigen</td>
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<td>HBV</td>
<td>Hepatitis B Virus</td>
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<td>HCV</td>
<td>Hepatitis C Virus</td>
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<td>HIV</td>
<td>Human Immunodeficiency Virus</td>
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<td>IEDCR</td>
<td>Institute of Epidemiology Disease Control and Research</td>
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<td>NAT</td>
<td>Nucleic Acid Amplification Technology</td>
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<tr>
<td>OFID</td>
<td>OPEC Fund for International Development</td>
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<td>RNA</td>
<td>Ribonucleic Acid</td>
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<td>RPR</td>
<td>Rapid Plasma Reagin</td>
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<tr>
<td>RR</td>
<td>Repeatedly Reactive</td>
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<td>SBTP</td>
<td>Safe Blood Transfusion Programme</td>
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<tr>
<td>SOP</td>
<td>Standard Operating Procedure</td>
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<tr>
<td>SRO</td>
<td>Statutory Regulation Order</td>
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<tr>
<td>TPHA</td>
<td>Treponema Pallidum Haemagglutination Assay</td>
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<tr>
<td>TTI</td>
<td>Transfusion Transmissible Infection</td>
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<tr>
<td>VDRL</td>
<td>Venereal Disease Research Laboratory</td>
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<td>WB</td>
<td>Western Blot</td>
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1. Introduction

A safe, effective blood transfusion service (BTS) is an essential component in the provision of an adequate health care service. It is the responsibility of a country to ensure safe blood by improvement of the service through accommodating modern facilities and techniques, establishment of infrastructure, development of manpower, and policies, and a National Blood Programme.

The Government of Bangladesh is committed to providing all patients with access to safe, sufficient and appropriate blood and blood products whenever needed. Recognizing the consequences of transfusing unsafe blood and components, only units which are negative on testing, for HIV, Hepatitis B and C, and other transfusion transmissible infections (TTIs), may be used. The Government also recognizes that safe blood is always in limited supply. The efficient coordination of the BTS at national level is a prerequisite for an effective and sustainable national blood screening programme. It is also required for the uniform application of national standards and procedures across the country. Coordination is essential in maintaining continuity in operations and consistency in performance in all facilities in which screening is performed, including blood centres and hospital based services. Each screening facility requires a specific and sufficient budget, a suitable infrastructure, with reliable water and power supplies, well-maintained equipment and efficient transportation and telecommunications systems.

During the time of the last two sector programmes under health services technical assistance, the major activities undertaken were capacity building, strengthening blood transfusion centres, developing blood component facilities, and conducting quality testing. Since 2000, there has been a gradual increment in the demand for blood due to improved and expanded health facilities, and it is estimated that 600,000 units of blood are required annually in Bangladesh. All donated units of blood in the country must be screened for HIV, Hepatitis B and C, syphilis and malaria as per the Safe Blood Act 2002.

The objective of blood screening is to detect infection markers in order to prevent the release of infected blood and blood components for clinical use. The assays selected for blood screening should be highly sensitive and specific. The aim is to detect all possibly infected donations while minimizing wastage due to false positive results. Donations which are reactive, or which give indeterminate test results, should be discarded using methods in accordance with standard safety precautions. Since the implementation of the SBTP there have been many issues about blood screening, including a basic understanding between diagnostic tests and screening tests, strategies for emergency screening and repeat testing, and the merits and demerits of pre- and post-donation screening. In addition, there is a need for confirmatory testing when an initial result is reactive, the management of positive blood donors, donor counselling, permanent deferral or reinstating of blood donors, selection of screening assays, interpretation of results of screening, steps in the blood screening process and screening algorithms.

All these points are illustrated in this document for clarification. Principles and strategies are derived mainly from WHO blood screening strategies that elaborate on issues related to blood screening in the blood transfusion services of Bangladesh. The current document was drafted to present recent strategies recommended by WHO for blood screening with interpretation of different strategies and processes to assist the national blood programme in standardizing blood donor management. The majority of references in this draft quote the WHO publication: “Screening Donated Blood for Transfusion Transmissible Infections” or the Draft National Blood Policy, Standard and SRO-145 Bangladesh, and recommendations of experts of the National BTS in Bangladesh.
1.1 Existing methodology for donor blood screening in Bangladesh

This mandates that all whole blood and apheresis donations must be tested for TTIs according to the Safe Blood Act which includes HIV, HBV, HCV, syphilis and malaria. Confirmatory testing in multiple small centres would lead to the wastage of precious resources and a lack of uniformity in standards. Therefore, it is also stipulated that one referral centre or regional centres must be identified by the national authority to perform confirmatory testing.

Mandatory screening began under the SBTP in 205 blood transfusion centres. The centres are integrated with medical college hospitals, institutes, combined military hospitals, specialized hospitals, district hospitals and the Upazila health complex. The strengthening of blood transfusion laboratories began under the SBTP in 2000. In the private sector, blood transfusion centres, either integrated into the hospital or stand-alone, operate under license to the Director General of Health Services. To date, there are 60 such blood centres operating in the private sector. These centres supply blood after screening for TTI markers to HIV, Hepatitis B, Hepatitis C, syphilis and malaria, the requirement of which has been made mandatory in accordance with the “Safe Blood Transfusion Law” in the country. According to a consolidated report of donor blood screening data from 205 centres, the prevalence of TTI markers in 2010 was <1%. Almost 98% of public centres use rapid assays for screening. This is a critical issue in evaluating the quality of blood screening in a decentralized system with more than one centre. Periodical external assessment of the prevalence of TTI markers is an effective tool. The implementation of Standard Operating Procedures (SOPs) for the processes involved in blood screening is also an effective tool in improving the safety of the blood supply.

The centres enlisted under the national SBTP intermittently receive support in the form of kits and reagents. However, because supply is not regular, some centres buy kits locally according to need. The current system of blood screening in each centre is therefore not regulated and controlled by the national external quality scheme and poses a risk to the quality of blood products; maintaining standards in a decentralized system is difficult.

- A needs assessment and feasibility of consolidating screening activities at national and/or regional levels may be considered in order for a national screening programme to be implemented – leading to the establishment of regional blood screening centres that operate more efficiently and cost-effectively. Additionally, a national screening programme that separates the activities of blood screening and clinical transfusion practice i.e. replaces the existing system in Bangladesh, would protect against product liability and focus on consumer protection. Standardization of suitable assays needs to be considered in addition to current rapid assays in referral/ regional centres, and identified testing centres should be strengthened with facilities for enzyme immunoassay (EIA), chemiluminescent immunoassay (CLIA) and Western Blot (WP) methodologies to support small centres like district and Upazila health complexes.
- Needs assessment should be done to identify requirements and priority interventions to strengthen TTI screening of donated blood and the network of facilities equipped to screen blood samples.
- Plans should include a monitoring and evaluation mechanism, with a baseline, targets and indicators in order to measure progress and impact on all facilities in which TTI screening of donated blood is performed.
1.2 National strategy for screening donated blood

Blood screening strategies have not been defined in Bangladesh, neither in SRO-145 nor in the Safe Blood Act. Efforts should be made to develop strategies by consensus, with clear decisions made on the scope of TTI screening as part of the blood screening programme. Strategies need to be appropriate to the specific situation, based on incidence and prevalence of infection, capacity and infrastructure of the BTS, the cost of screening and availability of resources. Whichever strategy is selected, it is important that it is implemented effectively and consistently within a well-managed quality system.

As per WHO guidelines, the national screening strategy should consider the following:

- Country specific infection marker(s) for which blood units must be tested
- Quality systems within which screening is to be performed
- Detailed description of screening methodology
  - Assay(s) to be used for each marker
- Algorithms to determine outcome of screening, i.e. fate of blood units tested
  - Interpretation of the results of screening tests including definition of initially reactive and non-reactive blood donations and decision points for the release of non-reactive units of whole blood and blood components
  - Whether initially reactive tests should be repeated or initially reactive donations should be discarded; the inclusion of repeat testing in the screening strategy as determined by the effectiveness of the quality system in place. The fate of initially reactive donations, non-reactive on repeat testing
  - Procedures for the quarantine and release or discard of blood and blood components
  - Whether, for donor management, confirmatory testing should be performed to distinguish between true reactivity and non-specific reactivity
- Blood screening in specific situations; for example, in remote areas with low workloads and limited facilities, when equipment is lacking or where there may be no electricity
- Emergency screening when blood is needed urgently
- The subsequent actions to be taken for donors whose blood tests are repeatedly reactive (RR), but are not confirmed positive: i.e. whether donors should be notified and counselled concerning possible non-specific or biologically false reactive results
- Donor look back and recipient follow-up
- Safe disposal of reactive and positive units

In order to implement the strategy, the SBTP should have:

- A system for the education and recruitment of low-risk voluntary non-remunerated blood donors
- National criteria for blood donor selection and deferral (which is already in place and available)
- Annual report on incidence and prevalence of specific infections in the general population and in blood donors. SBTP to combine and update reports from all public and private centres
- Suitable screening assays as per category of blood transfusion centre
- Suitable screening algorithm for each TTI
- Specific and sufficient budget for management of local hospital transfusion committees
- Suitable infrastructure, facilities and equipment, especially in regional centres and institutes, considering the introduction of EIA and CLIA
- Adequate and reliable supply of good quality test kits and reagents, according to an appropriate annual procurement plan. Assessment of the requirement for kits and consumables and a supply chain in place. Kits procured from preferred manufacturers whose kits are validated by the
IEDCR or other national or referral laboratory prior to delivery. Validation facilities developed in regional centres and discrepancies identified, reported and addressed

- Support for a national reference laboratory or access to such services
- Sufficient manpower with appropriate skills and competencies
- Facilities for confirmatory testing, donor counselling and a system for donor referral
2. Blood Screening Strategies

Strategy 1
In addition to blood group serology requirements, blood and blood components may not be released into stock unless they test negative in mandatory TTI screening for HBsAg, anti-HIV, anti-HCV, syphilis and malaria antibodies or antigen.

Strategy 2
Screening of donated blood should be carried out on samples taken during the donation process in a quality controlled environment.

Strategy 3
When stock of blood is limited and when there is a high prevalence of TTI markers, pre-donation screening may be applied. National guidelines should be developed for pre- and post-donation blood screening.

Strategy 4
All tests on blood samples should be performed and recorded in accordance with standardized procedures in laboratories that are properly equipped to undertake them.

Strategy 5
All blood samples and donations must be correctly labelled and identified throughout the screening process, in order for the correct results to be allocated to each unit; preventing errors and the transfusion of unsafe blood into a recipient.

Every blood transfusion centre ensures that separate storage equipment is clearly designated for:
- Unscreend units in quarantine
- Reactive/positive units
- Unresolved/indeterminate units
- Units suitable for clinical use, available blood stock

The expected performance standard of assays must be consistently achieved at testing sites, by using appropriate pre-acceptance batch testing and statistical monitoring of test results on defined quality control samples.

Strategy 6
Screening tests are to be performed with controls using validated procedures and SOP and documented accordingly. Results are recorded. Appropriate evaluation is done when selecting the type of assay for each TTI, based on critical assay characteristics, such as sensitivity and specificity, as well as cost and ease of use.

Strategy 7
After evaluation and validation of kits, specific algorithms should be initiated. A national screening algorithm is needed for each TTI for which testing is carried out.

Strategy 8
The assay selected for blood screening should be highly sensitive and specific. The aim is to detect all possibly infected donations whilst minimizing wastage due to false positive results. Donations that yield reactive or indeterminate test results should be discarded using methods in accordance with standard safety precautions.
Strategy 9
EIA and CLIA for blood screening are the assays of choice as they are suited to screening from relatively small to large numbers of samples.

Strategy 10
Rapid tests may be considered in the current context for use in blood transfusion centres with limited resources, and where only a limited number of tests are performed daily, e.g. district blood centres and Upazila health complexes.

The blood transfusion department of medical colleges, institutes and specialized hospitals should perform EIAs and CLIAs for blood screening in addition to the current system of rapid testing.

Strategy 11
The blood transfusion department of medical colleges as well as the regional blood transfusion centre should be provided more advanced TTI assays, so that reactive TTI results can be confirmed for blood donor notification and management. These centres should act as referral centres for other district and Upazila health complexes under their jurisdiction.

Strategy 12
After selection of a suitable blood donor, a seronegative result using a single (rapid/EIA/CLIA) test for TTI markers is sufficient to release the unit of donated blood for transfusion as per WHO and UNAIDS strategy for donor blood screening.

If a blood donation is found reactive after a single test (rapid/EIA/CLIA), the donated unit and any blood products derived from it must be segregated while the reactive result is investigated, either at the blood centre if re-testing facilities are available, or transferred to a larger centre for re-testing.

If a reactive blood sample is positive after re-testing at the blood centre, the confirmatory testing algorithm is followed as per national strategies for blood donor notification. The unit and all products derived from it must be discarded as per biosafety procedure. In blood centres where facilities for confirmatory TTI testing are not available, referral must be made to a medical college blood transfusion department/ regional blood centre for confirmation, donor notification and management.

Strategy 13
In emergency situations where blood and blood components are needed urgently, but are not available from the blood inventory, screening with rapid/simple single-use assays could be used to obtain results quickly and enable blood to be released for clinical use, in consultation with prescribing clinicians.

2.1 Types of assays for blood screening

The assays commonly in use are designed to detect antibodies, antigens or the nucleic acid of the infectious agent. However, not all assays are suitable in all situations; each has its limitations which need to be understood and taken into consideration when making a selection.

The main types of assay used for blood screening are:
- Immunoassays (IAs):
- Enzyme immunoassays (EIAs)
- Chemiluminescent immunoassays (CLIAs)
- Rapid/simple single-use assays (rapid tests)
- Nucleic acid amplification technology (NAT) assays.
In the context of blood screening, appropriate evaluation is required in selecting the type of assay for each TTI, based on critical assay characteristics, such as sensitivity and specificity, as well as cost and ease of use. Some description is given to assist in considering which assay(s) to adopt. However, there is no uniform strategy to accommodate all areas; the overall consolidation of screening assays needs to be considered in relation to the infrastructure of different types or categories of blood centres.

2.1.1 Immunoassays

Commonly used antigen detection assays are based on the use of immobilized antibody to capture pathogen-specific antigens present in the sample. Immunoassays can be used in different situations from high through-put laboratories with full automation to medium-sized laboratories with semi-automation to small laboratories, such as those in remote areas, which conduct a small number of tests manually.

2.1.2 Enzyme and chemiluminescent immunoassays

Enzyme immunoassays (EIAs) and chemiluminescent immunoassays (CLIAs) are currently the most commonly used assays for screening donated blood for TTIs. The design of EIAs and CLIAs is similar and they differ only in the mode of detection of immune complexes formed – colour generation in EIAs and measuring light produced by a chemical reaction in CLIAs. Any of these types of IA with high sensitivity will generally detect the target markers of infection required if they have been properly evaluated for blood screening and are then used within a quality environment. EIAs and CLIAs are suitable for the screening of large numbers of samples and require a range of specific equipment. These assays may be performed either manually or on non-dedicated automated assay processing systems (open system) or specific dedicated automated systems (closed system). EIA and CLIA have different solid phases to immobilize the antigen or antibody.

2.1.3 Rapid/simple single-use assays (rapid tests)

Rapid/simple single-use assays are discrete, individual, disposable assays: i.e. they are used once and discarded. These assays exist in a number of different presentations. Many rapid tests are based on a form of immunochromatography in which the added sample flows down an inert strip and reacts with previously immobilized reagents. The sample can be serum, plasma or even whole blood in some cases. Any positive reaction is visualized as a dot or a band appearing on the device strip. Most of the assays also include a control dot or band that is used to validate the results of each individual device, irrespective of the specific test result. Rapid tests are provided in simple-to-use formats that generally require no additional reagents except those supplied in the test kit. They are read visually and give a simple qualitative result within minutes. The reading of results is dependent on subjective evaluation and no permanent record of the original test results can be kept. Rapid tests are generally not suitable for screening large numbers of blood samples.

2.1.4 Nucleic acid amplification technology assays

Nucleic acid amplification technology (NAT), as applied to blood screening, detects the presence of viral nucleic acid, DNA or RNA, in donation samples. In this technology, a specific RNA/DNA segment of the virus is targeted and amplified in vitro. The amplification step enables the detection of low levels of virus in the original sample by increasing the amount of specific target present to a level that is easily detectable. The presence of specific nucleic acid indicates the presence of the virus itself and that the donation is likely to be infectious. NAT assays can either be performed on individual donations or on mini-pools to detect the nucleic acid of the infectious
agent. In addition to NAT assays which target individual viral nucleic acids, multiplex NAT screening assays have been developed which can detect DNA or RNA from multiple viruses simultaneously.

2.2 Selection of assays

The selection of appropriate assays is a critical part of the screening programme. Reliable results depend on the consistent use of well-validated and effective assays. A number of factors need to be considered in selecting the most appropriate assays. In general, a balance has to be found between screening needs and the resources available, including finances, staff and their expertise, equipment, consumables and disposables. Each screening system has its advantages and limitations that should be taken into consideration when selecting assays.

Advantages and limitations for consideration include:

- The length of time following infection before the screening test becomes reactive (window period).
- Rates of biological false positives which may result in the wastage of donations and unnecessary deferral of donors.
- The complexity of some systems that require automation.
- In most situations, EIA and CLIA assays developed specifically for blood screening are the assays of choice as they are suited to screening from relatively small to large numbers of samples. Most EIAs and CLIA have greater sensitivity and specificity than particle agglutination assays or rapid tests. Their manufacture and performance are generally more reliable and consistent and have better outcomes for blood screening.
- The use of rapid/simple assays is generally not recommended for blood screening as they are designed for the immediate and rapid testing of small numbers of samples, mainly for diagnostic purposes. These assays are performed using manual techniques; the results therefore have to be transcribed by staff and there is a lack of permanent records and traceability. As a result they may have limited use in blood centres, but are better suited to areas where blood inventory is low and only few donors are available, and where resources are limited with only a small number of tests performed daily. Rapid/ simple assays may therefore be useful in small district and Upazila hospitals, as they provide flexibility and no major pieces of equipment are needed.
- Rapid testing may be appropriate when a blood transfusion centre needs to screen specific donations on an emergency basis for immediate release of products due to a critically low blood inventory (i.e. in district and Upazila hospitals) or when rare blood is required urgently.
- In such emergency situations, the use of the rapid/ simple assays should be followed up with repeat testing using EIA or CLIA. These assays are routinely used at medical colleges and institutes.
- The introduction of NAT should be considered only when an efficient and effective programme based on antibody/ antigen testing is in place and there is a clear, evidence-based, additional benefit.
- NAT reduces the window period of infection. In countries with a low prevalence/ incidence of infection, the incremental gain is minimal as the number of donors in the window period at the time of donation is generally very low.

2.3 Assay characteristics

- Sensitivity and specificity are the key factors to be considered in selecting a specific assay.
- For the screening of blood donations, both sensitivity and specificity should be the highest available.
• Each assay should be evaluated within the country or region to confirm the technical data provided with regard to assay performance and, where possible, data from other studies should be analysed.
• Each assay should be validated in its place of use to assure that the performance is as expected according to the results of evaluation.

2.4 Evaluation of assays

• Assays produced by the major international diagnostics companies are generally well designed and are normally evaluated scientifically, both by the manufacturers themselves and by independent laboratories, prior to release onto the market.
• WHO operates a prequalification of diagnostics programme which provides technical information and advice on the quality of currently available HIV, malaria and hepatitis B and C test kits and assay systems with the aim of increasing access to affordable diagnostic technologies of assured quality that are appropriate for use in resource-limited settings.
• Data published in kit package inserts and the scientific literature also provide useful information, guiding selection of vendors, testing platforms and specific assays.
• Evaluations should be carried out in at least one major facility, but some BTSs may not have the necessary resources, expertise, experience and, importantly, panels of samples required. In such situations, the evaluations should be undertaken on behalf of the BTS, and in close conjunction with it, by an appropriate laboratory, such as a national reference laboratory.
• If none is available, the evaluation data required should be obtained from a BTS or reference laboratory in another country with similar demography, infection incidence and prevalence and BTS requirements, preferably in the same region. Reference should also be made to information available from laboratories elsewhere in the region, or globally.
3. Infection Markers for Blood Screening

3.1 Hepatitis B

HBV is an enveloped DNA virus transmitted via the parenteral route and may be found in blood and other body fluids. The virus travels to the liver where it replicates in hepatocytes. HBV is present in the bloodstream; the levels of the virus itself are variable. In recently infected individuals, viral DNA is normally present, although not always at high levels. Chronically infected individuals may either be infectious (viral DNA present) or non-infectious (viral DNA absent). Screening for hepatitis B surface antigen (HBsAg) indicates infection with HBV, but does not in itself distinguish between recent and chronic infections. The distinction between acute and chronic infection is not relevant to blood screening. HBsAg positive donations should be considered to be at high risk of transmitting HBV and should not be released for transfusion. When the HBsAg screen is negative, some individuals may have low levels of detectable viral DNA, which is transmissible by blood, and may therefore cause infection in the recipient.

Screening for Hepatitis B

During the course of infection, various serological markers develop, including HBsAg and hepatitis B core antibody (anti-HBc). In addition, HBV DNA can be detected in the majority of cases, although in HBsAg negative phases of infection the DNA levels are generally relatively low and the viraemia may be transient. The methods used to identify the presence of HBV employ the following screening targets:

- The detection of HBV DNA further reduces the risk of HBV transmission through the transfusion of infected blood donated during the acute window period: i.e. when the results of HBsAg assays are not yet positive, HBV DNA is positive.
- Low levels of HBV DNA have also been detected in the blood of individuals after the resolution of acute HBV infection and the disappearance of HBsAg, or in chronic occult HBV infection.

Markers for Hepatitis B screening

- HBsAg
- Anti-HBc
- HBV DNA

Hepatitis B surface antigen

- HBsAg is the prime marker used in blood screening programmes.
- It normally appears within three weeks of the first appearance of HBV DNA and levels rise rapidly.
- It can thus be detected easily by most of the highly sensitive HBsAg assays available.
- The presence of HBsAg may indicate current or chronic infection and thus potential infectivity.

Hepatitis B core antibody

- Anti-HBc is produced later in acute infection, after the appearance of HBsAg, and marks the start of the immune response to HBV infection.
- In general, anti-HBc persists for life, irrespective of whether the infection resolves or progresses to chronicity.
- In the vast majority of cases of Hepatitis B, the detection of anti-HBc has limited value as HBsAg is already present. In some cases, however, during the resolution of the infection, HBsAg may decline to below detectable levels. Although anti-HBs usually then appears relatively rapidly, there may be a short period of time prior to its appearance when anti-HBc is
the only detectable circulating serological marker of infection, even though the individual may still have low viraemia and would thus be potentially infectious.

To minimize the risk of HBV infection via transfusion

- Screening should be performed using a highly sensitive and specific HBsAg immunoassay (EIA/CLIA).
- Screening using a highly sensitive and specific HBsAg rapid assay or particle agglutination assay may be performed in laboratories with small throughput, in remote areas or in emergency situations.
- Screening for HBsAg indicates infection with HBV, but does not in itself distinguish between recent and chronic infections.

3.2 Hepatitis C

HCV is an enveloped RNA virus. It is transmissible by the parenteral route and may be found in blood and other body fluids. The virus travels to the liver where it replicates in hepatocytes, resulting in a similar picture to that seen with HBV infection. The loss of circulating antibody may leave no readily detectable evidence of previous infection. Several genotypes have been identified and are associated with different geographical distributions and some differences in immunogenicity and clinical features, including response to treatment with interferon alpha (IFN-α).

- Screening for both HCV antigen and antibody does not in itself distinguish between recent and chronic infection. The distinction is, however, not relevant to the screening of blood for transfusion and all HCV antigen and/or antibody positive donations are considered at high risk of transmitting HCV.

Markers for HCV screening

- Anti-HCV
- HCV antigen
- HCV RNA

HCV antibody and antigen

- HCV antibody becomes detectable approximately 30-60 days after infection.
- Viral antigen normally appears between 0-20 days after viral RNA first appears.
- Antibody is generated and can be detected 10-40 days after antigen is first detected.
- Serological screening has been highly effective in significantly reducing the transmission of HCV via transfusion.
- Until recently, anti-HCV has been the prime serological marker for blood screening programmes.
- HCV antigen can be detected in the peripheral blood earlier than antibody in the course of early infection.

Hepatitis C viral RNA

- Viral RNA is normally detectable within a few weeks of infection and persists for 6-8 weeks prior to antibody seroconversion.
- The detection of HCV RNA may further reduce the risk of HCV transmission by infected blood donated during the window period of antigen and antibody assays. When HCV antigen-antibody assays are negative due to the window period, HCV RNA is positive.
- Anti-HCV and HCV RNA tests have been introduced in some countries to improve the overall effectiveness of serological HCV screening.
To minimize the risk of HCV infection via transfusion

- Screening should be performed using a highly sensitive and specific anti-HCV immunoassay or a combination HCV antigen-antibody (EIA/CLIA) assay. The assay should be capable of detecting genotypes specific to the country or region.
- Screening using a highly sensitive and specific anti-HCV rapid assay may be performed in laboratories with small throughput, in remote areas or emergency situations.

3.3 Malaria

Malaria is caused by parasites of the *Plasmodium* species. Four main types infect humans: *P. falciparum*, *P. vivax*, *P. malariae* and *P. ovale*. Malaria is primarily transmitted to humans through the bite of a female anopheles mosquito. Although always of concern in endemic countries, malaria is increasingly a matter of concern to BTSs in non-endemic areas. Significant numbers of blood donors from non-endemic countries travel regularly to malarial areas and there is wide migration from endemic areas to non-endemic areas where migrants may then become donors. Malaria is gradually spreading into non-endemic areas or regions where it had previously been eradicated.

Although primarily transmitted by mosquitoes, malaria is readily transmitted by blood transfusion through donations collected from asymptomatic parasitaemic donors. The parasite is released into the bloodstream during its lifecycle and will therefore be present in blood donated by infected individuals. The parasites are stable in plasma and whole blood for at least 18 days when stored at +4°C and for extended periods in the frozen state.

Markers for malaria screening

The screening method may depend on whether it is endemic in the country or not. Methods used to identify the presence of malaria employ the following:

- Direct detection of parasite by thick film
- Serological markers: antibody and antigen

Endemic areas

In endemic areas, direct detection of parasites by thick film is often used to identify parasitaemic donations. However, the technique is time-consuming, highly operator-dependent and prone to error. High quality, sensitive malaria antigen assays are now readily available and may be better able to identify parasitaemic donations, including those with much lower levels of parasites than are reliably detectable by thick film. However, in endemic regions, if screening is even considered, screening strategies are generally complex, combining specific criteria for donor selection and deferral, based on the season, geography and availability of anti-malarial prophylaxis, with laboratory-based screening.

- Donor selection criteria should be developed to identify and collect blood from donors at the lowest risk of infection both during the malaria season and during the rest of the year.
- Donor selection and deferral strategies should be implemented to identify those donors with a current history of malaria or a specific identifiable exposure risk, such as travel to malarial areas, in which case the donor should be deferred for a period of three months.

Non-endemic areas

In non-endemic areas, the detection of specific antibody is effective for the screening of donations from individuals identified to be at risk of transmitting malaria. In virtually all cases, the deferral of risk individuals for a period of 3 years from the date of the last potential exposure, combined with malarial antibody testing, will prevent the transmission of malaria.
3.4 Syphilis

Syphilis is a sexually transmitted infection caused by the spirochete bacterium *Treponema pallidum*. Syphilis was one of the first recognized infectious risks of blood transfusion. The primary route of transmission is through sexual contact; it may also be transmitted from mother to fetus during pregnancy or at birth, resulting in congenital syphilis. Once in the bloodstream, the spirochetes spread throughout the body. A primary lesion, or chancre, usually occurs about three weeks after exposure, although the duration may be shorter in cases of transfusion transmitted infection where the organism enters the bloodstream directly.

- The treponemes are relatively fragile, in particular being heat-sensitive; storage below +20°C for more than 72 hours results in irreparable damage to the organism such that it is no longer infectious.
- Thus, although clearly potentially infectious, the risk of transmission through the transfusion of blood and blood components stored below +20°C is very low.
- Blood components stored at higher temperatures (above +20°C), such as platelet concentrates, or those not stored at lower temperatures for any length of time, such as blood collected and used within 48 hours, present a significantly higher risk of transmitting syphilis.

**Markers for syphilis screening**
- Non-specific, non-treponemal markers: antibody to lipoidal antigen (reagin)
- Specific treponemal antibodies

**Specific assays**
- Specific assays commonly used for blood screening are *Treponema pallidum* haemagglutination assays (TPHA) and EIAs. These detect specific treponemal antibodies and thus identify donations from anyone who has ever been infected with syphilis, whether recently or in the past, and whether treated or not.

**Non-specific assays**
- Non-specific assays such as Venereal Disease Research Laboratory (VDRL) and rapid plasma reagin (RPR) tests identify those individuals who may have been more recently infected.
- They detect antibodies to cardiolipin or lipoidal antigen (reagin); the plasma levels of these antibodies rise significantly in active infection due to cellular damage.
- The use of non-specific assays is of most value in diagnostic testing to identify recently infected individuals.
- When the incidence and prevalence of syphilis in the blood donor population are high and cannot be reduced through donor selection strategies, it may be necessary to consider screening using a non-treponemal assay (e.g. VDRL or RPR) to identify only the highest-risk donors – those with evidence of recent infections.

**To minimize the risk of syphilis infection via transfusion**

Screening should be performed using a highly sensitive and specific test for treponemal antibodies, either TPHA or EIA, supported by a non-specific test, i.e. VDRL or RPR.

3.5 Human Immunodeficiency Virus

HIV is a retrovirus, an enveloped RNA virus, which is transmissible by the parenteral route. The virus primarily infects and replicates in lymphocytes. The viral nucleic acid persists by integrating into the host cell DNA. HIV-1 and HIV-2 are the two major distinct virus types and there is significant cross-reactivity between them. HIV-1 is now endemic in many parts of the world,
although its incidence and prevalence is low in some regions. HIV-1 group M is responsible for more than 99% of infections worldwide, whereas the prevalence of HIV-2 is mainly restricted to countries in West Africa and India. The appearance of antibody marks the onset and persistence of infection but not immunity. HIV can be present in the bloodstream in high concentrations and is stable at temperatures at which blood and individual blood components are stored. The virus may be present in all products made from donated blood from an HIV-infected individual. Infectivity estimates for the transfusion of infected blood products are much higher (around 95%) than for other modes of HIV transmission owing to the much larger viral dose per exposure than for other routes. Screening donations for both antibody and antigen will identify the majority of donations from infected donors.

Markers for HIV screening
- Anti-HIV-1, including group O, + anti-HIV-2
- HIV p24 antigen (p24 Ag)
- Viral nucleic acid: HIV RNA

Anti-HIV-1 + anti-HIV-2 and p24 antigen
- Antibody screening strategies are the most commonly used; antigen screening methods should also be employed.
- Antibody may be detected approximately 3 weeks after infection and approximately 6 days after antigen is first detected.
- HIV p24 antigen may appear from 3-10 days after the appearance of viral RNA, and its detection can further reduce the serological window period by 3-7 days before antibody detection.
- Due to partial cross-reactivity only, between the main virus types (HIV-1 and HIV-2), it is not sufficient to rely on an HIV-1 specific assay to detect all cases of HIV-2.
- Use of antibody-only assays has been superseded by the use of combination HIV antigen and antibody assays (combined p24 and anti-HIV-1+2), wherever possible. This reduces the serological window period in early infection over antibody-only assays.
- HIV RNA can be detected approximately 7-11 days after infection: i.e. when the results of HIV antigen-antibody assays are negative, but HIV RNA is positive.
- The detection of HIV RNA can reduce the risk of HIV being transmitted through the transfusion of infected blood donated during the serological window period of antigen and antibody assays.

To minimize the risk of HIV infection via transfusion
- Screening should be performed using highly sensitive and specific test kits.
- Assay using anti-HIV-1+2 or HIV combination antigen-antibody test.
- Immunoassay (EIA/CLIA).
- Screening using a highly sensitive and specific anti-HIV-1+2 rapid assay may be performed in laboratories with small throughput, in remote areas or emergency situations.

Emergency screening
In emergency situations in which blood and blood components are needed urgently but are not readily available from blood inventory, screening with rapid/ simple single-use assays could be used to obtain results quickly and enable blood to be released for clinical use in consultation with the prescribing clinician.
4. Approaches to Blood Screening

Two approaches to blood screening are recommended for blood safety, depending on whether or not an effective quality system has been established in the testing laboratory. District hospitals and the Upazila health complex lack sufficient facilities, infrastructure and manpower for the establishment of a quality system. Even in larger and more advanced blood centres, the situation or environment to support a quality system may be inadequate. Considering these challenges to quality, several options are described and recommended for laboratories engaged in screening blood for TTIs:

- Quality systems are weak or have not yet been established
- Effective quality systems are in place

As per WHO recommendation the approaches are as follows:

4.1  In laboratories without well-established quality systems

These laboratories lack EIA and CLIA facilities, validated rapid devices kits, national/ international EQAS, experts/ trained manpower, facilities to store reactive samples for validation and quality control, parallel internal controls for periodical, regular checks on blood screening results.

- Use a single assay (e.g. called ‘A’) and test each blood sample singly in accordance with SOPs. The condition is that the assay used must have been validated for the specific TTI. This is particularly important if the kit has no record of successful validation or recommendation by the national referral laboratory or referral centres as recommended by the national expert committee. If the kits are supplied from the SBTP, the centre should be supplied with a written copy as evidence that validation was done and the kit found suitable for use. If the centre uses kits procured by local management, validation must be done prior to use, in a nearby regional centre, e.g. medical college or a centre recommended by the Safe Blood Authority.

- Collate and analyse the TTI results of the assay used:
  - If the result is non-reactive (i.e. A-), the blood unit maybe released for clinical use and issued to a patient for transfusion
  - If the result is reactive (i.e. A+), immediately separate the unit and then discard it

The decision not to use a reactive donation is taken on the basis of a single test strategy by using a single device. However, when a sample is found reactive, steps should be taken to exclude technical error or mix-up of samples. Therefore, the test should be repeated in duplicate (i.e. using two strips or devices or cards from the same manufacturer as that of the initial test) in the following way:

- Take the same sample from the same sample container and use the same kit, OR
- Take a sample from the tubing attached to the blood donation, and test the sample using the same kit

If the repeat result is non-reactive the unit cannot be released for transfusion as the laboratory lacks the criteria for quality assurance as described above.

The objective of repeating the test is to determine whether the individual performing the test followed the SOP, e.g. was the sample taken from the correct pilot tubing, were all the steps in the procedure carried out, were there errors during sample preparation, or when dispensing reagent
or sample, or when selecting of the device for TTI, or when using buffers, was the allocated time allowed for the development of results and the observance of the colour code of devices? This verification process is important; unless a thorough investigation is done corrective action cannot be taken to prevent the release of an unsafe unit of blood.

4.2 In laboratories with established quality systems

These laboratories have EIA, CLIA or other devices and facilities, have validated rapid device kits, are incorporated under regular EQAS nationally or internationally, have an adequate number of experts or trained manpower, have facilities to store reactive samples for validation and quality control, and the centre periodically and regularly runs its own controls on blood screening results, in parallel.

- Use a single assay (e.g. called ‘A’) and test each blood sample singly in accordance with SOPs. The condition is that the assay used must have been validated for the specific TTI. The centre has the capacity to validate kits before use, whether supplied from the SBTP or locally purchased by the centre.
- Collate and analyse the TTI results of the assay used:
  - If the result is non-reactive (i.e. A-), the blood unit may be released for clinical use and issued to a patient for transfusion.
  - If the result is reactive (i.e. A+), immediately separate and isolate the unit and all components derived from it.
  - Repeat the test in duplicate, from the same sample and using the same assay.
  - Analyse the results of the repeat duplicate tests:

If both repeat tests are non-reactive (A+, A-, A-) the initial result could be due to false reactivity, and the donation can be released for clinical use because the centre has established quality systems that fulfil all the criteria as stated above. However, this strategy needs precaution considering the prevalence of infection markers, national policy and also the strength of the quality system in the centre.

If one or both of the repeat tests are reactive (A+, A+, A+) immediately segregate and then discard the blood donation and all blood components derived from it. Send the sample for confirmatory testing.

4.3 Reference laboratory

Currently, almost all medical colleges and institutes in the public sector screen blood by using rapid assays. There have been a number of debates regarding this and it presents a challenge for the establishment of a reference centre under the National SBTP. A national public health/reference laboratory is generally suitable for this work. Alternatively, the role of the reference laboratory may be delegated to a BTS laboratory if it has suitable facilities, adequate resources and an effective quality system. An assessment of requirements for strengthening the reference laboratory may be needed to ensure its capacity to support the blood screening programme. The role of the reference laboratory may include:

- Evaluation and selection of assay systems and equipment
- Confirmatory testing on screen reactive donations for blood donor management
- Provision of quality control samples
- Organization of EQAS
Figure 1: Model Algorithm for Blood Screening

Non-reactive (A-)

Release donation and derived blood components

Perform initial screening test (A)

Initial reactive (A+)

Option 1
No/limited quality system
Discard donation and derived blood components

Option 2
Effective quality system
Repeat test in duplicate using same sample and same assay

Negative in both repeat tests (A+, A-, A-)
Release donation and derived blood components

Reactive in one or both repeat tests
(A+, A+, A-) / (A+, A-, A+) / (A+, A+, A+)
Discard donation and derived blood components
Send for confirmatory testing
5. Screening Algorithms

A screening algorithm sets out a sequence of steps in the blood screening process to be followed in each facility to determine the suitability of each unit of donated blood and its components for clinical or manufacturing use. It specifies the actual tests to be used and, based on each test result, directs the user to the next step. It defines the actual testing process. It includes clear statements on:

- Definition of screen non-reactive and decision points for the release of screen non-reactive donations
- Whether initially reactive tests should be repeated
- Fate of screen reactive donations
- Subsequent action to be taken with screen reactive donors

The use of screening algorithms helps to provide consistency in screening tests. It enables decision-making regarding the release of screened blood and blood components, the discard of unsuitable units and the management of blood donors with confirmed positive screening results.

A screening algorithm should be developed for each TTI. The algorithm is determined by the specific infection marker for screening, the expertise of users, the infrastructure, testing conditions and quality systems of individual screening facilities. Once an algorithm has been defined, this will guide the procurement of the specific test kits, reagents and laboratory testing systems required.

The algorithm must be validated for each TTI for which testing is done. An algorithm is the sequence of testing and selection of assays on which the final result (of testing for a TTI marker) is determined in a given strategy. The outcome must be validated in the context of blood screening in the country, and must be correlated with sensitivity and specificity, and with positive and negative predictive values relating to the prevalence of each TTI marker. For example, if the initial marker was reactive for HIV by ‘x’ type of assay, what type of assay would be selected in step 2 to determine whether step 1 was a true positive or negative, and what would the final tie-breaker assay be, to resolve the outcome if the result of step 2 was inconclusive?

The type of assay used for each step must be validated. Once this validation protocol is complete then the national authority finalizes and selects the types of assay for each step of the algorithm so that each centre follows the same algorithm. The sequence and timeline for adopting an algorithm, which must be pilot tested prior to approval, is as follows:

5.1 Key steps in developing an algorithm

- Identify appropriate tests
- Develop algorithm
- Build consensus
- Develop policy
- Expand to national scale
- Review testing algorithms annually
Table 1: Objectives of Testing

<table>
<thead>
<tr>
<th>Objective of Testing</th>
<th>Prevalence of Infection</th>
<th>Testing Strategy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Transfusion/ transplant safety</td>
<td>All prevalence</td>
<td>I</td>
</tr>
<tr>
<td>Surveillance</td>
<td>&gt;10%</td>
<td>I</td>
</tr>
<tr>
<td></td>
<td>&lt;10%</td>
<td>II</td>
</tr>
<tr>
<td>Diagnosis: clinical signs and symptoms of HIV infection</td>
<td>&gt;30%</td>
<td>I</td>
</tr>
<tr>
<td></td>
<td>&lt;30%</td>
<td>II</td>
</tr>
<tr>
<td>Asymptomatic</td>
<td>&gt;10%</td>
<td>II</td>
</tr>
<tr>
<td></td>
<td>&lt;10%</td>
<td>III</td>
</tr>
</tbody>
</table>

**Strategy I:** Single test is sufficient (e.g. to accept or reject a unit of donated blood)

**Strategy II:** Double test is required for surveillance or diagnosis

**Strategy III:** Triple test is required for diagnostic purposes

**Sensitivity**
- It is the accuracy with which a test can confirm the presence of an infection
- Tests with high sensitivity show few false negative results and are meant to screen blood prior to transfusion to maximize blood safety. However, very sensitive tests are more likely to include false positive results

**Specificity**
- It is the accuracy with which a test can confirm the absence of an infection
- Tests with high specificity show few false positive results and are to be preferred for the diagnosis of infection in an individual

**Figure 2: Timeline for Developing a National Testing Algorithm**

<table>
<thead>
<tr>
<th>Project Development</th>
<th>Test Evaluation / Algorithm Development</th>
<th>Monitoring</th>
</tr>
</thead>
<tbody>
<tr>
<td>Phase I</td>
<td>Phase II</td>
<td>Phase III</td>
</tr>
<tr>
<td>(~ 3 months)</td>
<td>(3-6 months)</td>
<td>(&gt; 3 months)</td>
</tr>
</tbody>
</table>

- Determine Capacity
- Literature Review
- Situation Analysis
- Needs Analysis
- Proposal
- Ethical Review Procurement
- Establish Panels
- Evaluation Analysis of Data
- Algorithm Decision
- Publish Findings
- Site Selection Training of Staff on Site Evaluation
- Algorithm Approval
- Pilot Manuals
- Monitor Performance
- Publish Algorithm
5.2 Blood Screening and diagnostic testing

Unless otherwise indicated, it is strongly recommended that screening on an apparently healthy individual is done as a single test. Samples from patients to be tested for pathology are referred to as being contaminated and requiring diagnosis, and must not be handled in blood centres, but in the hospital setting. This may lead to confusion when attempting to differentiate between screening and diagnostic scenarios. Table 2 below shows comparisons between these two situations.

Table 2: Comparison between blood screening and diagnostic testing

<table>
<thead>
<tr>
<th>No.</th>
<th>Conditions</th>
<th>Screening test</th>
<th>Diagnostic test</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Purpose of test</td>
<td>Determine presence or absence of infection marker in potential blood donor</td>
<td>Diagnosis in a symptomatic or asymptomatic individual being tested for health status</td>
</tr>
<tr>
<td>2</td>
<td>Target population</td>
<td>Healthy population</td>
<td>Potentially infected population</td>
</tr>
<tr>
<td>3</td>
<td>Action taken after testing</td>
<td>Selected for blood donation or deferred</td>
<td>Diagnosis of infection or follow up</td>
</tr>
<tr>
<td>4</td>
<td>Number of tests</td>
<td>Action taken after single test. Blood product related</td>
<td>Additional testing over a period of time. Case related</td>
</tr>
<tr>
<td>5</td>
<td>Additional test</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>6</td>
<td>Nature of sample</td>
<td>Low risk</td>
<td>High risk</td>
</tr>
</tbody>
</table>

5.3 Pre- and post-donation screening (testing)

In a small transfusion centre where the blood stock accumulates as a result of directed or family donations, donors usually prefer their screening test to be done before actually donating blood. The general understanding is that pre-donation testing for TTI markers is cost-saving on blood
bags, which would be wasted if the screening results proved positive and the blood donated had to be discarded. On the other hand, blood centres may prefer to perform post-donation testing for quick through-put of donors in the collection area, when it is expected that few donations will prove to be reactive.

In the case of outdoor campaigns the only option is post-donation screening because of the large number of blood donors to be handled over a short period of time. There are advantages and disadvantages to both approaches. The decision for pre- or post-donation testing depends on the local situation, especially on the stock of blood in the blood bank, the attitude of the donors and the local consumables and reagents to minimize cost. However, in an area of low TTI prevalence, post-donation screening (testing) may suit donors. In pre-donation screening, TTI statistics are not recorded and the opportunity of compiling a comprehensive blood donor database is therefore lost.

In summary:
• Pre-donation testing is sometimes considered a cost-saving measure, particularly in high-prevalence situations and also in centres where blood stock is inadequate.
• Pre-donation testing increases the time taken for a donor to donate blood, causing undue inconvenience to donors, and also the risk of discrimination and stigmatization.
• Pre-donation testing of donors does not ascertain the infectious status of the donation which requires further follow up tests on the blood sample collected during the blood donation process.
• Repeated venepuncture for first time blood donors may create a negative impact on the donation experience and discourage new donors.
• Post-donation screening, unlike pre-donation testing, is more comfortable for donors and reduces the workload on blood transfusion staff.
• Pre-donation testing may lead to wastage of resources and increased screening costs unless prevalence is high.
• The practice of pre-donation testing could undermine the long-term development of a sustainable blood donor programme based on carefully selected, voluntary non-remunerated blood donors who donate regularly.
• In an effective national blood screening programme, pre-donation testing of blood donors has limited applicability.
• In settings where the prevalence of an infection is high and donor selection would not be effective in reducing prevalence in first-time donors, pre-donation testing may be useful as an interim strategy while developing a stable pool of regular, voluntary non-remunerated donors.

5.4 Quarantine of blood and components prior to release or discard

It is recommended that before laboratory testing and screening for TTIs, a quarantine system should be in place for the physical segregation of all unscreened donations and their components. It is also recommended that a system should be in place to ensure that screened and unscreened units are stored in separate blood bank refrigerators to prevent the accidental issue of unscreened units. All reactive or positive donations and all components derived from these donations should be labelled “Not for transfusion” and segregated for discard or non-clinical use.

Each centre should ensure separate blood storage for:
• Unscreened units
• Reactive/ positive units
• Unresolved/ indeterminate units
• Units suitable for clinical use
Records of all storage equipment for blood and components must be maintained in a documented inventory system that identifies the current location and eventual fate of each unit of blood or component, whether destined for clinical use or disposal. All reactive units should be removed from quarantined stock and stored separately and securely until further handling or discard. Reactive or positive units of blood or plasma are valuable resources for quality control samples and panels, for evaluations and validations, and for research purposes, or to be used as reagents for quality assessment schemes or for the production of proficiency panels.
6. Confirmatory testing and blood donor management

In the course of blood screening, truly infected donors need to be identified, counselled and referred for medical care. There are debates on whether blood donors should have their reactive status confirmed at the centre where screening is carried out, or not. Most transfusion centres do not support a service for confirmation of blood donors due to a lack of facilities even in the transfusion department of medical colleges and institutes. Most commonly, initial reactive blood donors are deferred from blood transfusion centres and the donor is referred to another laboratory. This leads to confusion between donor and transfusion staff when the same donor presents with a non-reactive result from one centre and a reactive result that led to rejection in another. Therefore, confirmation of a blood donor is necessary for the appropriate follow up action to be taken.

Confirmatory testing should be performed by a reference laboratory or regional centre unless considerable expertise and resources are available within the BTS itself. The model algorithm shown in Figure 1 represents the minimum processes recommended for blood donor management and epidemiological monitoring based on initial screening and confirmatory testing. When blood screening is carried out in a laboratory with no (or limited) quality system, option 1 is followed, which states that reactive donations shall be discarded. Option 2 is followed where an effective quality system is in place, which states that blood from an initially reactive blood sample can be used when, after duplicate testing, both tests are non-reactive. The model algorithm shows the decision points on whether the donor should be accepted, counselled, deferred or referred, based on the results of confirmatory testing.

The confirmatory process also has an important role to play in public health as the close contacts of infected donors need to be protected from becoming infected. Confirmatory testing is an essential component of look back for ascertaining the true infectious status of the donor and recipients of previous donations. It also provides further benefit to the BTS in the epidemiological monitoring of infection rates in blood donors, thus contributing to a better understanding of donor behaviour and assessment of risk. Knowing and understanding confirmed infection rates in blood donors helps to ensure that donor selection, donor deferral and blood screening strategies are up-to-date and effective.

6.1 Interpretation and use of confirmatory results

Confirmatory testing is primarily concerned with the status of the donor and the subsequent action to be taken. Donations that are RR may be confirmed as being of negative, inconclusive or positive status.

- **A negative conclusion on confirmatory testing** indicates that the donor is not infected with the specific infection. However, a donor showing RR results on screening and negative results on confirmatory testing should be counselled and temporarily deferred until screening is non-reactive on follow-up. The donor can then be accepted for a donation.

- **An inconclusive outcome** is usually due to non-specific reactivity not related to the presence of the infectious agent. The donor should be counselled, deferred from blood donation and followed up for further investigations. In low incidence or prevalence countries, a significant proportion of blood donors whose donations give reactive screening results are not truly infected. A considerable number of donors may be lost due to deferral resulting from non-specific reactivity, especially if a test is not highly specific.
• **A positive conclusion** confirms that the donor is infected and should be permanently deferred from future blood donation, counselled and referred for appropriate medical care.

**Figure 4: Model Algorithm for Blood Donor Management**

6.2 **Managing blood donors**

The management of blood donors is an essential part of the activities of every BTS. Donors are very important because they are the source of the blood and blood components that are processed and released for clinical or manufacturing use. They should be managed in a way that ensures high standards of care and assures them of the concern of the BTS for their health and well-being. Blood screening and confirmatory testing enable the identification of infected donors or donors with non-specific reactivity or inconclusive results.

Even if only limited facilities are available, the BTS has a duty of care to donors, their families and the general population to ensure that infected individuals are referred for appropriate counselling,
treatment and further management as they may infect other individuals if they are not aware of their status.

The BTS and relevant authorities should have a clear policy and systems for communicating with these donors and informing them of their status in order to minimize any risk of further transmission. Donors who test negative for TTIs should be encouraged to donate regularly and lead a low-risk lifestyle.

6.3 Deferral of blood donors

Confirmed positive donors
Donors who are confirmed positive should be permanently deferred from blood donation, notified of their infection status, counselled and referred for clinical management as soon as possible.

Repeat Reactive but confirmed negative donors
The handling of RR donors with non-specific reactivity is a critical part of a screening programme because the selection of suitable screening assays and the use of an appropriate screening algorithm can minimize the unnecessary deferral of donors and loss of donations. Donors with RR results on screening and negative results on confirmatory testing should be informed, reassured, counselled and temporarily deferred until non-reactive on follow-up using the same screening assay or a different assay. If negative, they can again be accepted as blood donors.

Inconclusive results
Donors with inconclusive results present challenges to BTSs and screening laboratories as their management is less clear than with confirmed positive or confirmed negative donors. It is important to decide whether they can be retained on the donor panel or should be deferred. It is advisable to inform, counsel and defer inconclusive donors temporarily, usually for up to six months before re-testing.

If the screen is non-reactive and confirmed negative on follow-up, they may be accepted as blood donors in the future, subject to continued non-reactivity.

6.4 Reinstatement of donors

Where an initial sample taken at donation, and tested at a blood centre, is found RR, materials from that donation may not be used for transfusion, the donor’s record must be flagged in accordance with SOPs and the donor removed from the active panel. No further material from the donor must be used for clinical purposes until the donor has been returned to the active panel. Reinstatement may take place only if the donor’s follow-up sample tests negative on confirmatory testing and was therefore falsely reactive in the initial microbiology assay.

A specimen of the RR sample must be sent for confirmatory testing at a designated reference laboratory. If the specimen is considered to have been falsely reactive, reinstatement may be considered after a period of time. At least 12 weeks must elapse from the date of the first sample before the donor can be re-tested for consideration for reinstatement.

The specimen, taken at least 12 weeks after the initial bleed, must be sent to a designated reference laboratory. If the sample is now non-reactive in the current screening assay at the blood centre, and confirmed negative at the designated reference laboratory, the donor may be returned to the active panel as eligible for future donations. When the next donation is given, it may be used only if a negative result is obtained in the screening test.
7. Post-donation counselling

Informing donors that they are confirmed positive for an infection clearly poses sensitive issues and donors need to be counselled on the results and the actions that should subsequently be taken. Where feasible, the BTS should appoint a specialist donor counselling officer, trained to conduct counselling interviews, and provide referrals to agencies for further counselling, treatment and care. If applicable, the officer should ask the donor to provide contact details for his/her physician, who is then able to contact the patient and communicate directly.

Informing donors of non-specific reactivity is highly problematical and should be undertaken with care because this reactivity often varies and usually does not have an impact on the physical health of the individuals.

The permanent deferral of these donors is sometimes considered to be unnecessary, but may be unavoidable unless policies and procedures are in place that recognize variable non-specific reactivity and facilitate the appropriate management of such donors.

Post-donation counselling of donors can provide information on the possible routes of infection and the effectiveness of donor education and donor selection criteria, including why the donors decided to donate, whether they already knew they were infected and whether donor education materials gave sufficient information about risk behaviour. This kind of information aids in understanding patterns of infection in healthy individuals and can be used to ensure that donor information and education materials are clear and unambiguous. They can be used to improve donor selection criteria and the donor selection process.
8. Look back and recall

The look back procedure involves identifying the recipients of a donation which tested negative for a particular microbial marker and which tests positive on the next donation.

One of the major hazards of blood transfusion is the transmission of TTIs. The retrospective procedure (look back) consists of tracing the recipients of the unit(s) of blood concerned, in order to determine whether or not infection was transmitted.

The blood and blood components, prepared and transfused from the previous donation, are traced to individual recipients who are then notified, counselled and tested for evidence of infection. Look back first became a routine practice with the introduction of HIV testing of blood donations. Look back for HIV was universally applied in the UK, and in the USA. Look back may identify recipients infected through possible “window period” donations.

Both donors and recipients need to be counselled and re-tested. The responsibility for counselling and testing of a recipient rests with the clinician managing the patient and the responsibility for tracing the donors, donations, and the re-testing and counselling of donors is the responsibility of the BTS.

Recipient of blood from seroconverting donor
Guidelines should be in place for counselling a patient who has received a transfusion of blood donated by a donor who, at a subsequent donation, tests positive for a TTI. The donor may have been infected after the previous donation and the recipient may not have been infected. However, it is possible that the donor was infected at the time of the donation in question, and tested negative due to being in the “window period”.

Pre-test counselling
- Reassure the patient that this is a routine procedure
- Question the recipient on risk behaviour other than blood transfusion e.g. sexual promiscuity, multiple sexual partners or sharing sharps and needles.
- Reassure the patient after taking a sample of blood for TTI screening

Post-test counselling
This counselling is done after the TTI screening has been completed. Depending on the system in place, this may take from 2 days to 2 weeks.