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Advances in pretransfusion infectious disease testing: ensuring the safety of transfusion therapy

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At the dawn of the third millennium, the American blood supply has never been safer. This is largely the result of advances in infectious disease testing for transfusion-transmitted viruses (TTV) that have occurred over the past 20 years [1–3]. Today, most transfusion-associated morbidity and mortality occur not from TTV infections, but rather from clerical errors or bacterial contamination of blood products.

Infectious disease testing of blood donations began with testing for syphilis (1940s), followed by testing for hepatitis B surface antigen (HBsAg) (1970); antibody to HIV (1985); antibody to hepatitis B core (anti-HBc) (1986); antibody to human T-cell lymphotropic virus (HTLV)–I (1988); antibody to hepatitis C virus (HCV) (1990); antibody to HIV-1 and -2 (1992); HIV-1 p24 antigen (1996); and antibody to HTLV-I and -II (1998). These serologic tests, the implementation of nucleic acid amplification testing (NAT) [4], and blood donor screening questions targeted at high-risk behavior have substantially decreased the risk for TTV [5–7]. The most recent estimated risks for TTV are for HIV-1, 1 in 1.9×10^6; for hepatitis B (HBV), 1 in 1.4×10^5; for HCV, 1 in 5.4×10^5 [8]; and for HTLV-I and -II, 1 in 6.3×10^5 [5].

Although over 99.99% of the time transfusions are safe from these viral agents [6,8], the public's perception of blood safety continues to be one of doubt, and its demand for a zero-risk blood supply persists. Recognition of transfusion transmission of HIV in the mid-1980s heightened sensitivity

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in the private, public, and governmental spheres to the potential risk of transfusing infectious agents and negatively transformed the public's perception of the safety of the American blood supply [7,9,10]. The evolution and appearance of previously unknown pathogens, such as new variant Creuztfeldt-Jakob disease (vCJD), that may or may not be transfusion-transmitted raise questions for regulatory agencies and the blood industry regarding the protection of the blood supply from unknown or emerging infectious agents [11].

This article reviews the required serologic pretransfusion infectious disease tests, discusses the advances in pretransfusion infectious disease testing using NAT, and describes research in the prevention of bacterial contamination of blood products [12,13] and pathogen inactivation strategies, all of which help to ensure the safety of the American blood supply.

Overview of serologic testing of donor blood

Since 1972 regulatory oversight of blood and blood products in the United States has been under the domain of the Food and Drug Administration (FDA) as detailed in Title 21 of the Code of Federal Regulations (Parts 600-799, 200-299, and 800-899) [14]. The FDA mandates which infectious disease tests are required for donor blood, and how they are to be performed. The FDA is rigorous in its control of the collection, testing, manufacturing, distribution, and transfusion of blood products. The FDA disseminates regulatory information and requirements to the blood industry, hospitals, physicians, and transfusion services through a series of publications. The Code of Federal Regulations is published annually. The FDA promulgates more timely information, such as new regulatory requirements or revisions of current regulations in memoranda, and also issues guidelines. FDA regulations set the minimum requirements for the operation of hospital transfusion services, blood banks, and blood collection centers. They are used by accrediting agencies, such as American Association of Blood Banks [15] and the College of American Pathologists, to formulate practice standards and for the content of accreditation inspections.

As of July 1, 2001, the FDA mandates that donor blood in the United States be tested for HBV, HCV, HTLV, HIV, and syphilis as follows: HBsAg, anti-HB core, anti-HCV, anti–HTLV-I, anti–HTLV-II, HIV p24 antigen, anti–HIV-1, anti–HIV-2, and a serologic test for syphilis. The testing facility must strictly adhere to the FDA-approved manufacturer's package inserts for each infectious disease test performed.

Pretransfusion serologic testing for TTV is based on detecting a viral-induced antibody or viral-associated antigen. To understand the different testing strategies for TTV, it is helpful to understand the dynamics of viral infections (Fig. 1). The time of entry of the pathogen into the host is called exposure. This is followed by a period of viral replication resulting in the detection of viral DNA or RNA, followed by detection of the viral antigen,

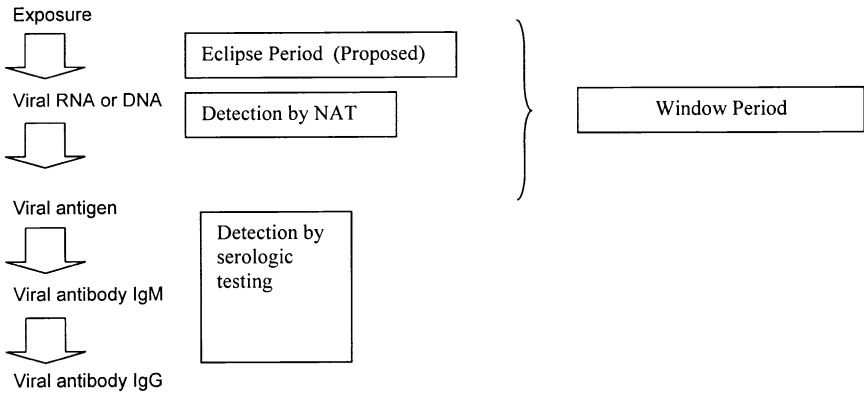

Fig. 1. Dynamics of viral infection.

and finally detection of the viral antibody. The time period between donor exposure to the virus, and the detection of viral DNA or RNA, has tentatively been called the eclipse period because it has been proposed that before the detection of viral DNA or RNA, the donor may be noninfectious [5]. The time periods between donor exposure to the virus, the detection of DNA or RNA, viral antigen, and viral antibody are specific to each virus. The time from viral exposure to the appearance of serologically detectable viral antigen or antibody is called the window period. The residual risk of TTV is primarily caused by donors in this window period of infection in whom TTV are undetected by serologic screening [5]. One of the ongoing goals of pretransfusion infectious disease testing has been to decrease the duration of the window period, thereby improve detection of TTV, and consequently improve the safety of the blood supply. This is the rationale for the development of nucleic acid–based tests.

The pretransfusion serologic testing algorithm is initiated with a group of screening tests for either the presence of viral antigen (HBsAg and HIV p24 antigen) or antibody to the virus (anti–HIV-1 and -2, anti-HCV, anti–HTLV-I and -II, and anti-HBc). A negative screening test is called nonreactive and the donor unit is approved for transfusion. If, however, a donor's blood sample tests positive for a TTV antigen or antibody, the sample is called initially reactive. The screening test is then repeated in duplicate on the same sample. If both of the repeat tests are negative, the donor sample is considered nonreactive, and the donor unit is acceptable for transfusion. If one or both of the repeat tests is positive, however, the sample is considered repeat reactive, and the donor unit is discarded. Repeat reactive samples are then tested with a confirmatory test or a supplementary test (if available). A confirmatory test is one that actually establishes that the analyte was present in the original test (Roger Dodd and Susan Stramer, personal communication, 2001). The blocking or inhibitory assays used to confirm HbsAg and HIV-1 p24 antigen and also for syphilis are confirmatory, because a specific antibody is used to

block reactivity in the confirmatory test. In contrast, a supplementary test is a different test that supports or refutes the original screening test. By this logic, the Western blot and related tests are supplementary. Although the confirmatory and supplementary tests have defined criteria for positive and negative results, variably the pattern manifests as neither positive nor negative, and is called indeterminate [16]. There is no confirmatory or supplementary test for donors who have repeat reactive anti-HBc.

Donor deferral decisions are based on repeat reactive results. FDA regulatory requirements do not allow further blood donation from the donor who has a repeat reactive screening test, unless and until the donor has completed a re-entry testing algorithm. After a repeat reactive screening test, the donor's eligibility for blood donation is put on hold as the positive test result is further evaluated. A donor with a repeat reactive viral screening test and a positive or indeterminate supplementary or confirmatory test is not eligible for reentry and is indefinitely deferred from blood donation. A donor with a repeat reactive screening test that is negative by additional confirmatory or supplemental testing may be evaluated for re-entry as a donor. A follow-up sample is drawn later ranging from 1 to 6 months [17] and if all infectious disease tests are negative at that time the donor is eligible to donate and may be reentered as a donor.

A positive confirmatory or supplementary test for HIV or HCV initiates the FDA-mandated "look-back" process. Look-back involves identifying all recipients who received seronegative or untested blood products from a donor who is later discovered to have a confirmatory or supplementary test positive for a TTV. The FDA has created time lines and standards for look-back process that are specific for each virus [17].

At times when specific tests for TTV were not available, surrogate testing also has been implemented in pretransfusion infectious disease testing. Alanine aminotransferase and anti-HBc are two surrogate tests added to pretransfusion infectious disease testing to help identify non-A non-B hepatitis, before the identification of HCV. The introduction of surrogate tests has been met with controversy because these tests are of limited specificity in the donor population [17].

Because the new testing methodology NAT adds sensitivity to infectious disease testing and is less prone to ambiguity in its interpretation and application for donor deferrals, it may better adjudicate decisions as to which donors will be permanently deferred.

Viral infections

Hepatitis B

The DNA *Hepadnavirus* (HBV) was the first known TTV. Currently, HBV is detected in donor blood by testing for HBsAg and anti-HBc. First

described in 1965 in Australian aborigines by Blumberg et al [18], the HBsAg particles are produced in excess during acute HBV infection and in the chronic carrier state. Infectivity with HBV may exist for up to 40 days before the HBsAg is detected. Current HBsAg testing methodologies detect 0.1 to 0.2 ng/mL of HBsAg or 3×10^7 viral particles. Transmission of HBV from HBsAg-seronegative donors, however, has been reported. A donor who tests positive for HBsAg may be either in the early phase of acute infection (without anti-HBc or with IgM anti-HBc) or is a chronic HBV carrier with IgG anti-HBc. Age at the time of infection is a risk factor for becoming a hepatitis B carrier with only 5% of adults becoming carriers postinfection, whereas 90% of infected children become chronic carriers. The clearance of HBsAg and appearance of anti-HBsAg mark the resolution of HBV infection. Anti-HBsAg is not included in blood donor screening because it is found in individuals who have been vaccinated against HBV. Anti-HBc usually appears after HBsAg is detected but before the start of clinical symptoms and persists for years following infection and also is a surrogate marker for HCV infection [19].

Hepatitis C

Formerly known as non-A non-B hepatitis, the RNA *Flavivirus* (HCV) was identified in 1989 [20]; by 1990 an antibody screening test for anti-HCV had been developed, implemented, and added to routine donor blood testing [21]. There is currently no approved test in the United States to detect anti-HCV antigen in blood donors. Anti-HCV is detected at approximately 10 weeks after infection with current anti-HCV screening tests. The significance of a positive screening test (anti-HCV) in an otherwise healthy blood donor is unclear without supplementary testing. In the United States between 0.5% and 1.4% of blood donors have repeat reactive anti-HCV screening tests. For donors with repeat reactive screening tests, supplementary testing with slot immunoblot assay is performed to determine the HCV antibody specificities. A positive supplementary test strongly correlates with infection with HCV.

Human retroviruses: HIV-1, HIV-2, HTLV-I, and HTLV-II

HIV type 1 (1983) was the first virus characterized as the causative agent of AIDS. To detect HIV-1, donor blood is tested for the HIV antigen p24 and for anti–HIV-1. Another strain of the virus, HIV-2, more commonly found in Africa and rare in the United States, was discovered later and in 1992 anti–HIV-2 was added as a required blood donor test. Most blood centers use a combination enzyme immunoassay, which detects anti–HIV-1 and anti–HIV-2, but leaves a 22- to 25-day window period open from the time of donor infection to the time of anti–HIV-1 and -2 detection. A negative screening test for anti–HIV-1 and -2 does not guarantee a transfusion free of HIV-1 and -2. For example, Ling et al [22] presented a case

of transmission of HIV-1 and the failure of routine anti–HIV-1 testing to detect the virus during the infectious window period. The addition of HIV-1 p24 antigen to donor blood testing in 1996 shortened the window period to 15 to 16 days.

The HTLV types I and II are *Oncornavirus* or *Oncoviruses* that may be transmitted through transfusion of cellular blood products. HTLV-I was the first human retrovirus isolated and has been associated with adult T-cell leukemia, tropical spastic paraparesis, and a myelopathy consisting of a semiprogressive neurologic disease [23]. Tropical spastic paraparesis is also known as *HTLV-associated myelopathy* and occurs in the minority of individuals infected with the virus (2% to 4%). HTLV-I has been shown to be transmissible by blood contact, sexual contact, through breast milk (vertical transmission), and most recently associated with allogeneic bone marrow transplantation [24]. HTLV-II was described after HTLV-I and is associated with large granular lymphocytic leukemia and leukopenic chronic T-cell leukemia. Screening for HTLV-I and HTLV-II is done on donor serum with an assay (combination HTLV-I and -II or HTLV-I cross-reactive) that tests for anti–HTLV-I and -II. Approximately two-thirds of initially reactive donor samples fail to react on repeat testing and only about 10% of the repeat reactive samples are later shown to be infectious [25]. Influenza immunization can cause false-positive screening tests for anti–HTLV-I and -II and is thought to be caused by nonspecific IgM cross-reactivity. The supplementary test for HTLV-I is Western blot [16].

Bacteria

Syphilis

The venereal disease-producing spirochete *Treponema pallidum* has been transmitted by blood transfusion. Although the spirochete does not survive at the 1°C to 6°C storage temperature of red cells, platelets stored at room temperature may transmit the organism. Screening tests for syphilis include the rapid plasma reagin (RPR) test and the hemagglutination test. The RPR detects the antibody-like substance reagin, which is directed against the widely distributed lipoidal antigen cardiolipin. These cardiolipin antibodies routinely develop in individuals after an infection with syphilis but also may occur with other entities. The RPR test is performed by placing donor serum on a card with cardiolipin-coated charcoal particles, which serve as visual indicator of antibody-antigen agglutination. The hemagglutination test is more specific for antibodies to *T pallidum*, and requires placing donor serum in microtiter plates filled with fixed chicken erythrocytes sensitized with components of *T pallidum*. The presence of antibodies resulting in hemagglutination is read photometrically, and may be automated. If either screening test is reactive, a confirmatory test specific for the spirochete is

performed, such as fluorescent treponemal antibody absorption. Both the screening and confirmatory tests for syphilis generate false-positive results in the blood donor population, which is a low-risk population [26]. The American Red Cross evaluated the change from a RPR test to an automated specific treponemal test (PK-TP) in screening for syphilis in blood donors and found that the change to the PK-TP test resulted in a lower repeatedly reactive rate, better prediction that a confirmed-positive test for syphilis occurs in testing in the fluorescent treponemal antibody absorption, fewer donations lost, and comparable deferral rates [27].

NAT

Nucleic acid amplification testing and technology (genome amplification testing) is the latest methodology in the battery of infectious disease tests for donor blood and uses genomic amplification strategies from molecular biology (polymerase chain reaction [PCR] or transcription-mediated amplification) to amplify viral DNA or RNA. NAT is performed on pools of donor plasma containing 16 to 24 donor samples. This represents a paradigm shift from the serologic testing model, which tests individual donor samples for infectious disease markers. Because of the genomic amplification technology, facilities must have a separate area designated for the amplification process to prevent contamination. The primary advantage of NAT is that it reduces the risk of infection from HCV and HIV-1 and -2 by identifying donors who have been infected by a virus, but have not yet made serologically detectable antibodies and are in the window period of infection [4,28–30]. This increased sensitivity is demonstrated in pooled donor samples for HIV and HCV, where viral loads early in infection are large. Increased sensitivity is not achieved on pooled samples for HBV, where sample dilution abrogates the potential increase in sensitivity [31].

Nucleic acid amplification testing was first implemented in Europe, where in Germany it has been a mandatory part of blood donor testing since April 1999. On February 28, 2002 the FDA licensed HIV and HCV NAT for screening donors of whole blood and blood components intended for use in transfusion. The approved test system was developed by GenProbe Inc., San Diego, California.

Nucleic acid amplification testing reduces the window period of viral detection for HBV, HCV, and HIV. NAT detects the HBV DNA at extremely low levels in donor plasma (unpooled only) before the donor is seropositive for HBsAg. In the chimpanzee model it is estimated that NAT detects as few as 10 genomic copies of the HBV DNA, which is equivalent to an infectious dose [32–34]. This low-level viremia that occurs before rapid viral replication coupled with the slow doubling rate of the virus (about every 3 days), however, makes the amount of HBV DNA present in the plasma much lower, and NAT, especially on pools of donor samples, seems to

be less useful in routine donor blood screening for HBV compared with HIV and HCV.

For HCV, NAT has reduced the length of window period from approximately 70 to 80 days using only anti-HCV testing to 10 to 30 days using anti-HCV and NAT [35,36]. In Germany, Hitzler and Runkel [37] implemented routine HCV RNA PCR in blood donor screening and tested 251,737 blood donations by HCV PCR. They found three (1:84,000) in which anti-HCV was not detected in the antibody screening test but was detected using PCR. Roth et al [31] in Germany found 2 of 370,000 donations to be HCV RNA positive and antibody screen negative in the first 2 years of testing. Roth later reported that from January 1997 through January 2000 his institution tested 1,078,940 donations in 13,274 pools and a total of three HCV PCR-only positives were identified [61]. In the United States in the first year of NAT for HCV the yield for HCV RNA confirmed-positive, seronegative donations was 62 HCV NAT-reactive donations among more than 16.3 million screened (1:263,000) [38]. Of the HCV NAT-reactive donations identified, one had not seroconverted over a 300-day period [38].

Nucleic acid amplification testing has decreased the window period for HIV from 24 days to 12 to 13 days and is superior to testing for p24 antigen [5,6,28]. In the first year of NAT testing in the United States the combined yield for HIV RNA confirmed-positive, seronegative donations was four HIV NAT-reactive, p24 antigen-negative donations among greater than 12.6 million screened (1:13,150,000) [38].

Nucleic acid amplification testing is rapidly evolving. Presently, NAT is performed on pooled plasma. Research is underway for automation of NAT for testing individual blood units [39]. Lee and Prince [39] recently presented a new method for automation of NAT using polyvinylidene fluoride filter plates. This method permits full automation of the simultaneous extraction of nucleic acids of HCV, HIV, and HBV from donor sera and permits NAT screening of individual units of blood. Standardization of NAT is necessary before introducing NAT in routine donor blood testing for HBV, HCV, and HIV [40]. In April 2001 the World Health Organization established a preparation as the first international standard for HIV-1 RNA for use in NAT testing [41].

Bacterial contamination

As advances in testing have reduced the risks of TTV, the risk of other infectious agents has proportionally increased. Despite rigorous and thorough screening questions and the use of aseptic technique at the time of phlebotomy, bacterial contamination of blood components occurs; transfusion of a contaminated unit may result in severe sepsis with mortality rates reported up to 26%. The true incidence of the problem of bacterial contamination is unknown, but certainly is underestimated [13].

Although the required temperatures used to store red blood cells, cryoprecipitate, and plasma inhibit bacterial growth, room temperature storage of platelets promotes the bacterial growth. Bacterial contamination accounted for 29 (16%) of 182 transfusion-associated mortalities reported to the FDA from 1986 to 1991 [42] and for 23% of transfusion-related fatalities reported to the French Blood Agency from 1994 to 1996 [43]. The incidence of bacterial contamination of platelets has been reported in as many as 1 of 524 random-donor pools [44]. Bacterial contamination research is currently focused on developing methods for bacterial diversion, detection, and destruction.

Diversion

The skin of the blood donor is the most common source of bacterial contamination. Phlebotomy needles can "core out" a skin plug and its bacteria-containing cutaneous adnexal structures, which are then carried with the first milliliters of blood collected into the bag [45]. One strategy to reduce this risk of contamination has been to develop a bag or collection technique where the first 15 to 30 mL collected is diverted from the unit [46]. This diverted blood is then conveniently used for the mandatory blood donor screening tests while reducing the risk of bacterial contamination.

Detection

Unlike the tests for TTV agents, which are performed on a sample obtained concurrently with the donation, bacteria may be present in numbers too small to be detectable at the time of phlebotomy. Bacteria detection strategies must take into account the time needed for bacterial proliferation and optimize the timing of bacterial detection in accordance with the bacterial growth cycle. Detection of bacterial RNA with antibody probes [47,49], measuring either glucose consumption or oxygen consumption (by the partial pressure of oxygen) [83], Gram staining [44], and automated culture systems [47,50,51] have been investigated to decrease bacterial contamination. Nucleic acid amplification and a chemiluminescence-linked RNA probe rapidly can detect in less than an hour 10^4 to 10^5 bacteria per milliliter. Bacterial metabolic processes consume glucose and produce organic acids and these changes can be used to detect bacteria. Burstain et al [48] documented decreased glucose and pH levels in stored platelets using chemistry dipsticks, although the technique was not reliable and yielded false-positive results. Aerobic bacteria consume oxygen; hence, the background oxygen compared with the partial pressure oxygen in bag can be compared to assess bacterial proliferation [83]. Finally, some research has focused on the pathogen inactivation as a means to destroy contaminated units. The most sensitive method at this time, achieving 10 CFU per mL, is microbiologic culture or specially designed pouches [52].

The two strategies, which achieved limited clinical application, are the Gram stain and culture. Yomtovian et al [44] sampled platelet units before issuance and performed a pretransfusion Gram stain, which was sensitive to 10^6 to 10^7 bacteria per milliliter. Using this strategy, several instances of contaminated platelets were interdicted before transfusion. Lack of sensitivity, however, allowed many other culture-positive (in retrospect) but Gram stain–negative units to be transfused with variable clinical sequelae. An increasingly popular detection strategy, pretransfusion culture, achieves sufficient sensitivity only after an approximately 2-day holding period [53,54]. Any widespread application of this strategy likely necessitates a concomitant increase in the platelet storage interval from 5 to 7 days. The other strategies mentioned are still under development and have not yet achieved clinical application.

Pathogen inactivation (destruction)

The goal of pathogen inactivation is to inactivate known pathogens including bacteria and potentially decrease the risk of transfusion-transmitted infectious agents [55]. In the United States solvent detergent plasma (SD plasma) is the only currently FDA-approved strategy for pathogen inactivation. Other areas of research include the experimental photochemical treatments of psoralen S-59, inactine, and riboflavin [56].

Pathogen inactivation technology for SD plasma involves pooling plasma in lot sizes of about 2500 units (donors) per pool, adding solvent 1% tri-n-butylphosphate and detergent 1% triton X-100, which dissolve the walls of lipid-enveloped viruses, such as HBV, HCV, HIV-1 and -2, HTLV-I and -II, and cytomegalovirus [57,58]. SD plasma is currently an ABO blood group–specific, pooled plasma product. Limitations of this technology include that it requires pooled plasma, which increases the number of donor exposures for the recipient; it fails to inactivate nonenveloped viruses, like hepatitis A and parvovirus B19 [59]; and it is not applicable to cellular components. In addition, because of deficient α_2-antiplasmin, it may promote bleeding in patients with serious hepatic disorders [60,61].

Photochemical inactivation of pathogens involves the addition of a chemical to the blood product, which binds to the pathogen's nucleic acids and is then activated by long wavelength ultraviolet light thereby interfering with nucleic acid function. Psoralen S-59, under clinical trials with fresh frozen plasma and platelet preparations, is a synthetic psoralen that intercalates to viral nucleic acid genomes; after exposure to ultraviolet radiation the genomes become cross-linked and are unable to replicate [62]. Psoralen S-59 does not seem to interfere with platelet function [63,64]. Psoralen S-59 and may be used on single units of fresh frozen plasma, which avoids the pooling risks of SD-plasma. It also blocks DNA replication in mononuclear cells and may help to reduce graft-versus-host reactions [65,66] and decrease cytokine production implicated in febrile transfusion reactions [67]. This

treatment inactivates some nonenveloped viruses (parvovirus B19), bacteria, and protozoa. It should inactivate unknown pathogens provided their nucleic acid genome is accessible to psoralen S-59 [68]. Psoralen S-59 is variably effective against gram-negative and -positive bacteria [69]. Inactine is another chemical currently in clinical trials for use with red blood cells that prevents DNA and RNA genome replication and may inactive bacteria, protozoa, and viruses with no apparent effect on erythrocyte function [70]. An alternative strategy exploits a natural chemical reaction between riboflavin (vitamin B_2) and light to inactivate pathogens in blood products. With this process, known as *pathogen eradication technology*, riboflavin is added to the blood component, which is then exposed to light during which an oxidative photochemical reaction occurs destroying DNA and RNA without harming erythrocyte and platelet function. Unlike psoralen and inactine, pathogen eradication technology does not require removal of potentially toxic psoralen by-products after treatment because vitamin B_2 is a naturally occurring compound and is readily metabolized by the body [56].

The future: fears and solutions?

An ongoing concern is that unknown or emerging infections will compromise blood safety. Before the HIV-AIDS epidemic such an eventuality was thought improbable but experience has taught otherwise. The emerging public fear of vCJD, the human counterpart of mad-cow disease, being possibly transmitted by transfusion reflects the impact of the unknown on scientific practice and decision-making [71].

Transmissible spongiform encephalopathies result from the accumulation in tissues of a pathologic isoform PrP^{Sc} of the normal cellular prion protein PrP (PrP^{c}) [72]. The transmissible spongiform encephalopathy in its classic form, CJD is a rapidly progressive, fatal neurodegenerative disease characterized by progressive dementia and motor disturbances [11,73,74]. Most cases are sporadic (85%) [75] and are caused by a proteinaceous particle termed a *prion*. The remaining cases are familial, caused by the inheritance of a mutation in the prion gene, which in its nonmutated form encodes for a normal cellular protein, or iatrogenic. The incidence of CJD in the United States is approximately 1 in 1,000,000. Fatal CJD has been transmitted through vectors that have contact with central nervous system tissue (growth hormone derived from human pituitary, allografting of dura mater, and insertion of contaminated intracerebral electrodes) causing concern about the possible secondary transmission of CJD through blood products and prompting regulatory agencies to defer from blood donation any individual with risk factors for CJD [76,77]. Most recently, vCJD has been diagnosed in over 100 people primarily in Great Britain and represents transmission from the agent of the bovine transmissible spongiform encephalopathy presumably by ingesting contaminated meat or items containing

rendered animal by-products. Both bovine spongiform encephalopathy and CJD are caused by prions that seem to resist inactivation by nucleases, ultraviolet irradiation, treatment with psoralens, boiling, or proteases, but that can be inactivated by sodium hydroxide [78]. Fear of the theoretic risk of transfusion transmission of vCJD in a subclinical carrier phase has prompted deferral policies for donors who have traveled to the United Kingdom and Europe for variable periods of time.

Research is being actively pursued to detect the vCJD PrP^{Sc} [79,80] in the blood. Although it has yet to be proved that this abnormal prion may be transmitted by blood transfusion, one very preliminary study has demonstrated transmission of vCJD in a single sheep transfused with blood from an infected sheep in the incubation period, heightening the fear of potential transfusion transmission in humans [81].

Summary

The public expects a zero-tolerance policy for the transmission of infectious agents by blood transfusion. Although unrealistic, the efforts to reach this goal have produced an extremely safe albeit costly blood supply [82]. Blood collecting agencies, the FDA, physicians, and scientists have over the past 20 years created a complex system of layers of protection to interdict transfusion-transmitted infections (Fig. 2). As new, exotic, potentially blood transmittable infectious agents evolve [83], new barriers will be erected to

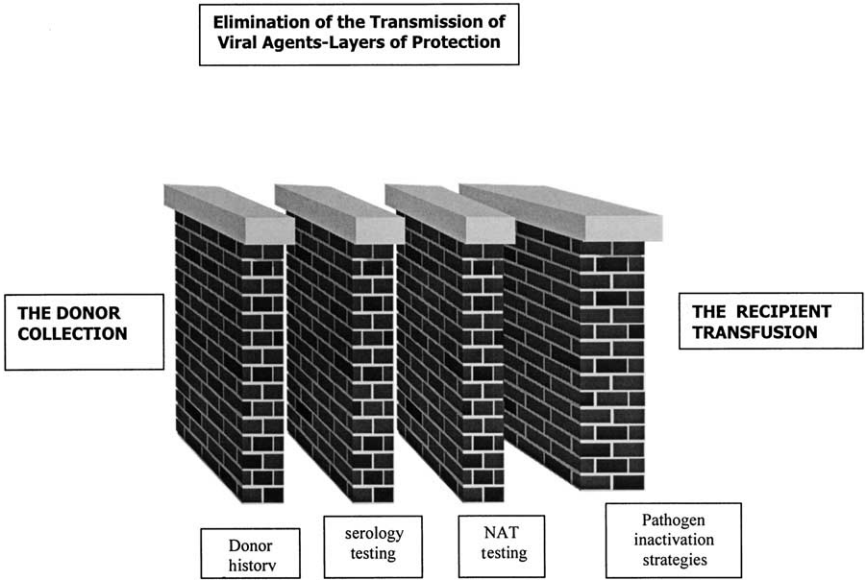

Fig. 2. Layers of protection.

interdict these agents. In the interim, the US blood supply is the safest in the world.

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