

Chapter 6: Donor Selection, Donor Testing and Pathogen Reduction

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BACKGROUND

All blood transfused in Canada is collected from volunteer donors. To ensure the safety of the blood products, donors are carefully screened against an extended list of eligibility criteria. In addition, the donated blood is tested to identify the donor's blood group and to detect blood group antibodies and transfusion-transmissible pathogens. Donor eligibility criteria and testing also benefit the donors by, for example, reducing potential blood donation-related health risks.

This chapter describes the donor selection process, the testing done on donated blood, and the pathogen inactivation processes that can further reduce the potential risk of transfusion-transmitted diseases. This chapter is complemented by [chapter 7](#) (Fractionated blood products and associated pathogen safety) and [chapter 8](#) (Pre-transfusion testing) of this Guide.

DONOR SELECTION

Donors are questioned about medical conditions and behaviors to determine if their blood donation would pose an increased risk to their own health or the health of the recipient.¹⁻³

Donors must provide identification at registration. Computer records are checked to determine whether a deferral code has been attributed to the donor after previous donations. Donors are asked to read a pamphlet ("[What you must know to give blood](#)") explaining the donation process, the testing that will be done on their blood and the obligatory provincial requirements for reporting certain test results to public health authorities. The pamphlet also explains the transfusion-transmission risk factors for human immunodeficiency virus (HIV) and hepatitis and informs donors that testing may fail to identify individuals who are in the early stages of infection.

After reading the pamphlet, the donor's medical history is assessed with a standard electronic questionnaire. Donors may fill out the questionnaire at home on the day of donation, or in the clinic. Donors are asked about illness in major organ systems that may put them at increased risk of an adverse reaction to donation. They are also asked about risk factors for transfusion-transmissible diseases. As laboratory tests have improved, the importance of the health assessment questionnaire in eliminating donors at risk for infectious diseases has decreased. However, currently, the questionnaire is the only means of excluding donors with a risk of Creutzfeldt-Jakob disease (CJD), variant CJD, Ebola virus, malaria, Zika virus, babesiosis or leishmaniasis. Testing is not performed for these agents.

Selective testing is performed for Chagas disease, on donors identified as being at risk based on the questionnaire. Selective testing is also performed for West Nile Virus (WNV) during the winter months on donors identified as being at risk based on the questionnaire. Donors are also asked about travel outside of Canada, the continental US and Europe, and deferred for three weeks after their return to reduce the risk of transmission of Zika virus. In addition, individuals taking teratogenic medications are identified and excluded from donation. Depending on the magnitude of the risk, donors may be deferred temporarily or indefinitely. For example, currently, people who have taken illegal drugs by injection are indefinitely deferred, while travellers to a region where malaria is considered endemic are deferred from donation of cellular blood components for one year.

Donors must be at least 17 years old (18 years old at Héma-Québec) and meet certain height and weight requirements. For younger donors, more stringent height and weight criteria may apply. Approximately 13% of donors are determined to be ineligible; most of these are temporary deferrals. It is estimated that less than four in 100 eligible Canadians donate blood each year. The average donor donates slightly less than twice a year.

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Chapter 6: Donor Selection, Donor Testing and Pathogen Reduction

Over one-third of deferrals are due to inadequate hemoglobin levels. Based on studies of iron deficiency in blood donors, Canadian Blood Services recently revised its whole blood donor eligibility criteria.⁴ Female donors may only donate whole blood every 84 days (maximum 4 times a year); the minimum inter-donation interval for male donors remains 56 days (maximum 7 times a year). The minimum hemoglobin level was increased to 130 g per litre for male donors; the minimum level remained at 125 g per litre for female donors. At Héma-Québec, the one difference is the minimum inter-donation interval for females which is 56 days. For more information, see our publication [The importance of iron for whole blood donors: a Canadian perspective](#).

BLOOD COLLECTION PROCESS

Whole Blood Donations

Prior to donation, the donor's temperature is taken and must be below 37.5 °C. In addition, a screening test for hemoglobin level is done on a capillary blood sample. Both arms are examined for signs of injection intravenous drug use. The donor's skin is disinfected using a swab stick containing 2% chlorhexidine gluconate and 70% isopropyl alcohol. An alternative disinfection protocol is used for donors allergic to chlorhexidine. For more information read our publication [Alternative methods of blood donor skin disinfection](#).

Phlebotomy is performed using a sterile single-use kit that contains an anticoagulant nutritive solution. The time for phlebotomy varies from 10 to 15 minutes. Approximately 480 ml of blood are collected per donation. The first few millilitres of blood are directed to a diversion pouch before filling the main collection bag. The diversion pouch has been shown to decrease the contamination of the collection bag with bacterial skin flora. The blood in the diversion pouch is used for the serological and infectious disease testing performed on each unit of blood collected.

Apheresis Donations

The process for screening apheresis donations is very similar to the one for whole blood donation. Several additional criteria are present to ensure the safety of the donor and the quality of the blood product.

Plateletpheresis donors must have a platelet count of 150×10^9 per litre prior to undergoing each procedure. Plateletpheresis may be performed every 14 days for a maximum of 24 donations in a calendar year. Depending on donor characteristics, such as initial platelet count and blood volume, each plateletpheresis donation may yield single or multiple platelet units, or both platelet and plasma products.

Plasmapheresis donors must have a total serum protein of over 60 g/l at each donation and a normal serum protein composition, verified periodically. Donors may make weekly plasmapheresis donations. The maximum quantity of plasma that may be collected per donation and during a twelve-month period is dependent on the donor's weight. Alternatively, the donor's estimated blood volume (EBV), based on sex, weight and height, may be used to calculate the maximum amount of plasma that can be collected.

SCREENING TESTS PERFORMED ON BLOOD DONATIONS

Transfusion-Transmissible Disease Testing

Multiple screening tests are performed on each donation to detect the presence of transfusion-transmissible infectious agents. The screening tests performed may detect antigens, antibodies or nucleic acids of the infectious agents. Table 1 provides the screening and confirmatory tests performed at Canadian Blood Services.

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Chapter 6: Donor Selection, Donor Testing and Pathogen Reduction

Antibody and antigen tests are done on individual donor samples while nucleic acid testing (NAT) is primarily done on pools of six samples.^{5, 6} The multiplex assay used for NAT enables the simultaneous detection of HIV RNA, hepatitis C virus (HCV) RNA and hepatitis B virus (HBV) DNA. West Nile Virus (WNV) RNA testing is also done in pools of six samples. However, to enhance sensitivity, single unit WNV NAT may be used in selected geographic areas during outbreaks of WNV. Apart from detecting both lineages of WNV, the WNV assay will also react with other viruses in the Japanese encephalitis virus serocomplex including Japanese encephalitis virus, St. Louis encephalitis virus, Murray Valley encephalitis virus, and Kunjin virus.

NAT for HIV, HCV and HBV are performed on all blood donations throughout the year. Testing for WNV is performed based on potential risk. During the WNV season, from June 1st to November 30th, all donations are tested for WNV due to the increased risk. However, during the winter months, from December 1st to May 31st, WNV testing is performed only on donors who are identified as being at-risk on the donor questionnaire because of travel outside of Canada in the eight weeks prior to their current donation.

Testing for antibodies to *Trypanosoma cruzi* (Chagas disease) is performed on donors identified as being at-risk on the donor questionnaire. Donors are designated as at-risk if they, their mother or their maternal grandmother was born in Mexico, Central America or South America or if the donor has resided in these at-risk countries for a significant period of time.^{7, 8}

Testing for antibodies to cytomegalovirus (CMV) is performed on a small subset of donations to provide CMV-negative products only for fetuses receiving intrauterine transfusions.⁹ Since CMV is a cell-associated virus and all cellular blood components are leukoreduced in Canada, the risk of CMV infection related to blood products is extremely low. For more information, see our publication [Use of cytomegalovirus \(CMV\) seronegative blood products](#).

Table 1: Transfusion-transmissible disease testing at Canadian Blood Services.

Pathogens	Screen Tests	Confirmatory/Supplemental Tests
HIV 1/2 (human immunodeficiency virus, types 1 and 2)	<ul style="list-style-type: none"> • Anti-HIV-1/2 HIV NAT 	<ul style="list-style-type: none"> • Geenius HIV-1/2 confirmatory testing
HBV (hepatitis B virus)	<ul style="list-style-type: none"> • Hepatitis B surface antigen (HBsAg) • Antibody to hepatitis B core antigen (HBcore) • HBV NAT 	<ul style="list-style-type: none"> • HBsAg confirmatory testing
HCV (hepatitis C virus)	<ul style="list-style-type: none"> • Anti-HCV • HCV NAT 	<ul style="list-style-type: none"> • HCV Line testing (LIA)
HTLV-I/II (human T-cell lymphotropic viruses, types I and II)	<ul style="list-style-type: none"> • Anti-HTLV I/II 	<ul style="list-style-type: none"> • HTLV Western Blot Assay testing

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Chapter 6: Donor Selection, Donor Testing and Pathogen Reduction

Pathogens	Screen Tests	Confirmatory/Supplemental Tests
<i>Treponema pallidum</i> (pathogen for syphilis)	<ul style="list-style-type: none"> • Micro-hemagglutination assay for <i>Treponema pallidum</i> (MHATP) 	<ul style="list-style-type: none"> • Algorithms may vary depending on reference laboratory site and initial/follow-up results • <i>Treponema pallidum</i> Particle Agglutination (TPPA) Test • Rapid Plasma Reagin (RPR) • Fluorescent treponema antibody absorption (FTA-ABS)
WNV (West Nile virus)*	<ul style="list-style-type: none"> • WNV NAT 	<ul style="list-style-type: none"> • Sequencing or alternate NAT may be done at reference laboratory on WNV NAT-positive specimens when history suggests a potential exposure to another member of Japanese encephalitis virus serocomplex
<i>Trypanosoma cruzi</i> (pathogen for Chagas)*	<ul style="list-style-type: none"> • Anti-<i>Trypanosoma cruzi</i> chemiluminescent assay 	<ul style="list-style-type: none"> • Enzyme-linked immunosorbent assay (ELISA) • Immunoblot • Polymerase chain reaction (PCR)
CMV (cytomegalovirus)†	<ul style="list-style-type: none"> • Anti-CMV particle agglutination assay 	<ul style="list-style-type: none"> • None available
<p>*performed on at-risk donors. †performed on a subset of donors for components required for IUT.</p>		

Donors who have initially reactive results on antigen or antibody testing have repeat testing performed twice on the same sample. If one of these two repeats is reactive, the donation is discarded, and additional testing is performed to determine the true status of the donor for donor notification and counselling. For some markers, donors may return and be re-tested at least six months post-donation, and resume donation if test results are negative (donor re-entry). Depending on the viral marker, inventory retrieval of blood products from previous donations and notification of the hospitals that received blood products from previous donations (lookback process) may be performed. For more information on this process see [chapter 1](#) of this Guide.

Nucleic Acid Amplification Testing (NAT)

For NAT multiplex screen testing, a reactive pool is resolved to an individual reactive specimen and the viral target that it is reactive for (HIV RNA, HCV RNA or HBV DNA). For WNV the reactive pool is resolved to the individual reactive specimen. Donors reactive on the multiplex screen test are indefinitely deferred, whereas donors reactive on the WNV screen test are deferred for 56 days.

Bacterial Testing

Because platelets are stored at room temperature for up to seven days, they are the blood product most likely to support bacterial growth.¹⁰ All platelet units are tested for bacterial contamination using an automated blood culture system incubated for up to six days after inoculation (eight days after collection). Platelets are issued as “negative to date”, and will be recalled, along with other blood products from the same donation, if the culture subsequently becomes positive.

Blood Group Determination and Antibody Detection

ABO, Rh D and K (Kell) typing are performed using an automated hemagglutination assay. Confirmatory typing is done for first-time donors found to be Rh negative, and donors are tested for the presence of Rh D and weak

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Chapter 6: Donor Selection, Donor Testing and Pathogen Reduction

D antigens. Canadian Blood Services implemented K testing on all donations in November 2018 and by the end of March 2019, nearly 90% of active donors had been phenotyped. The Kell phenotype appears on the end label if the donor is antigen negative. Testing is also performed for unexpected red blood cell antibodies. The methods used may be less sensitive than those required in pre-transfusion antibody detection. In recipient testing, a low level of antibody may be of clinical importance, since an anamnestic response may occur. In donor testing, only a small amount of passive antibody transfusion will occur and is generally clinically insignificant for red cell transfusion. Plasma and platelets are not produced from donations with red blood cell alloantibodies.

Phenotyping for RhC/c, RhE/e, Duffy, Kidd and S/s is routinely performed on a subset of repeat donor samples using an automated, algorithmic approach. This has led to a substantial inventory of red blood cell units end-labeled as negative for those antigens. The number of donors tested depends on the antigen. In general, approximately 30% of donors have been phenotyped for the common clinically significant antigens. Red blood cell antigen genotyping is also performed on some donors. This testing allows for identification of certain rare phenotypes and confirms variable serologic reactivity for some antigens. Genotype testing may also be performed when serologic reagents are not readily available to define a particular red blood cell antigen type or when family members of rare donors are investigated as part of the Canadian Blood Services rare blood donor program. Negative phenotypes defined by serologic or genotyping methods are reported on the end label. See our [Serological best practices](#) resources and our publication on [phenotype matching for sickle cell patients](#) for more information on how to use antigen typed blood products in transfusion practice.

Platelet products can be tested for human leukocyte antigen (HLA) or human platelet antigens (HPA) to manage patients who, for example, have documented antibodies targeting a HPA or alloimmune refractoriness. For more information about platelet products testing and utilization see [chapters 12](#) and [chapter 18](#) of this Guide.

PATHOGEN INACTIVATION SYSTEMS

Current pathogen inactivation (PI) technologies are based on chemical or photochemical principles that target and damage nucleic acids (disrupt DNA and RNA). They significantly limit replication and growth of pathogens such as bacteria, viruses and protozoa thereby preventing infectivity.^{11, 12} Certain PI technologies are marketed for the treatment of single transfusion doses of plasma or platelet products in some jurisdictions. Applications of these technologies for the treatment of red blood cell products and for units of whole blood are under development in clinical trials.^{11, 13}

PI methods for plasma products include solvent-detergent treatment of plasma in pools and methylene blue followed by ultraviolet (UV) A illumination for single unit plasma.¹⁴⁻¹⁶

Both these methods are in use in several European and other non-North American jurisdictions.^{17, 18}

PI methods for both plasma and platelets products include amotosalen S59 (a psoralen) plus UVA illumination, riboflavin plus UVA/B light illumination, and UVC illumination alone.^{11, 19} All three methods are in use for plasma and/or platelets products in Europe, Asia, the Middle East, Central and South America. Amotosalen plus UVA illumination was approved for apheresis platelets in the United States in 2015. Riboflavin plus UVA/B light illumination for platelet products was the subject of a recently completed international clinical trial in which Canada was a participant.^{20, 21}

Amotosalen plus UV illumination was licensed for platelet products in Canada in 2018. A pooled plasma product that has undergone PI using solvent-detergent is licensed and is available with some restrictions.

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CONTINUING PROFESSIONAL DEVELOPMENT CREDITS

Fellows and health-care professionals who participate in the Canadian Royal College's Maintenance of Certification (MOC) Program can claim the reading of the Clinical Guide to Transfusion as a continuing professional development (CPD) activity under Section 2: Self-learning credit. The reading of one chapter is equivalent to two credits.

We're here to answer your questions about the Clinical Guide to Transfusion. We'd also appreciate your ideas on how to improve the Guide. Please contact us through the [Clinical Guide feedback form](#).

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Chapter 6: Donor Selection, Donor Testing and Pathogen Reduction

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