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Procedure Summary

Environmental Health and Safety at West Texas A&M University (WTAMU) is composed of two distinct but integrated environmental safety departments which report to the Vice President of Research and Compliance. Academic and Research Environmental Health and Safety (AR-EHS) is responsible for research and academic related compliance, which includes laboratory and academic research and the associated compliance committees. Fire and Life Safety (FLS-EHS) is responsible for fire related compliance and conducts fire and life safety inspections of campus buildings and assists with the testing of all fire detection and suppression systems.

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Purpose

To provide standards for phlebotomy and retrieval of blood samples, in a manner which ensures the delivery of safe, competent, and effective care.

Blood Collection

Proper infection control and prevention practices are essential and are included at the end of this procedure manual (**Appendix 1**). Personal protective clothing (gloves, lab coats, etc.) and personal protective equipment (eyeglasses, safety goggles, full-face shield, etc) need to be worn at all times when blood is handled. According to OSHA, gloves must be worn whenever drawing blood for clinical testing without exception. However, OSHA does not specify when gloves must be put on. Gloveless palpation is allowed within the HPRL as long as the phlebotomist exercises proper hand hygiene between participants. Gloves must be worn on both hands when the vein is accessed. Once the venous site is chosen, proper cleansing must be completed; after that point, the person drawing the blood should not touch the site again with a bare or gloved hand.

Upon arrival, participants need to be greeted and identified. A brief description of this greeting and other questions you should ask the participant are included at the end of this procedure manual (**Appendix 2**). Following this greeting the participant should be directed to the dedicated phlebotomy station that contains: 1) a clean surface with two chairs (one for the phlebotomist and the other for the patient); 2) a hand wash basin with soap, running water and paper towels; and 3) alcohol hand rub. Note that a comfortable reclining couch with an arm rest is also available and it is up to the participant whether they choose to sit up or lie down. If the participant expresses a tendency to faint during blood collection procedures, the participant is highly encouraged to lie down during the procedure. To remove the risk of environmental contamination with pathogens, counter and work surfaces, and chair arms should be cleaned with disinfectant at the start of each shift and when visibly dirty.

The most common site for performing a venipuncture is the antecubital area of the arm because of the accessibility of three large veins: the median cubital or median, the cephalic and the basilic. In addition, most participants have six potential acceptable veins in their combined antecubital areas. In some, though, it can be difficult to locate even one. Age, obesity, genetics, dehydration, chemotherapy and many other medical conditions and treatments can be factors that make finding antecubital veins difficult. A figure depicting each of these potential venipuncture sites has been included at the end of this procedure manual (**Appendix 3**).

Not all phlebotomists have the confidence to puncture veins they can feel but cannot see. Those new to the procedure or those who perform venipunctures infrequently are often moved to consider alternative sites. As one's phlebotomy skills improve and they become more reliant on the sense of touch rather than sight, however, confidence in performing antecubital venipunctures increases. Because swelling makes locating veins more difficult and can prolong healing and closure of the puncture site, venipunctures should be avoided in an arm with edema. Additionally excessive swelling can alter the composition of the blood

collected from the affected limb. Naturally, venipunctures to an arm that is injured, burned, scarred, or otherwise traumatized should be avoided. This includes hematomas. Nor should infected, inflamed, or excessively bruised antecubital areas be considered. While it is unlikely that participants have had a mastectomy prior to venipuncture in our laboratory, it is critical that they are asked. If they answer “yes,” then NO venipunctures should be performed in the affected side’s arm. Significant lymph node removal often accompanies surgical mastectomy procedures. Because lymph nodes regulate fluid balance, their removal effectively interferes with lymph flow (lymphostasis) in the respective limb. In addition, blood collected from the same side as a mastectomy may contain higher concentrations of lymphocytes and waste products normally contained in the lymph fluid.

Begin by tightening a tourniquet 5 to 6 finger widths above the antecubital area. This tourniquet should be thoroughly disinfected with 70% isopropyl alcohol prior to each use. This practice ensures that infection control procedures are in place and the health and safety of participants is protected. Participant comfort is also important. The tourniquet should remain flat around the circumference of the upper arm, not roll into a rope-like constrictor. To avoid pinching the skin, tighten the tourniquet around a sleeve if possible. Alternatively, a blood pressure cuff can be used in place of a tourniquet if inflated to a level below the participant’s diastolic pressure or near 40 mm Hg is usually adequate. Tourniquets should be tight but not uncomfortable. Tuck a loop of the tourniquet between the tourniquet and the arm so as to provide an easy, one-handed release. After applying constriction, instruct the participant to clench the fist or grab the rubber ball. Discourage pumping of the fist as such activity can elevate levels of potassium and ionized calcium in the blood stream. Pumping the fist also brings about movement in the antecubital area that can interfere with vein location.

Locate the most prominent of the three acceptable veins of the antecubital area visually and by palpation by lightly pressing down on the skin repeatedly with varying degrees of pressure to detect underlying veins. Too much pressure may not allow for the tactile sensation of the vein’s curvature or elasticity. Likewise, too little pressure may not bring the finger close enough to feel the vein. The median cubital vein typically lies in the center of the antecubital area; the cephalic is on the outside (lateral) aspect; the basilic is on the inside (medial) aspect of the antecubital area. Veins will feel spongy, resilient, and have a tube-like curvature. Tendons and bone are harder structures and distinctively different. Attempt to locate the median cubital vein first because it is the vein of choice. The median cubital vein is only the vein of choice, however, if it is visible or palpable and if there is a high degree of confidence that it can be accessed successfully. Choosing another vein over the median cubital when the median cubital is clearly present can increase the risk for injury. However, selecting an indistinct median cubital vein over one of the other clearly visible or palpable veins may result in an unsuccessful attempt and subject the participant to a second puncture unnecessarily. If a median cubital vein cannot be located, repeat palpation on the lateral (outer) aspect of the arm where the cephalic vein lies. Complete the survey by palpating the skin on the medial (inner) aspect of the antecubital area where the basilic vein lies. Even if a cephalic or basilic vein has been located, repeat the survey on the opposite arm (if accessible)

in an attempt to locate the median cubital vein. Phlebotomists who select a basilic vein before surveying for a median cubital or cephalic vein do not reduce the risk of injury to the lowest possible degree. That's because two branches of the antebrachial cutaneous nerve lie in close proximity to this vein, sometimes passing between the skin and the vein itself. Once pierced, these nerves send shooting pain down the length of the limb to the fingers, sometimes up to the shoulder and into the chest. Nerve injury is often disabling and can be permanent. Most nerve injuries that result from punctures in the antecubital area occur during attempts to puncture the basilic vein. Since nerves are neither visible nor palpable; avoiding the basilic vein when other prominent veins are available reduces the risk of nerve injury. Attempt to locate the median cubital vein on either arm before considering alternative veins. Due to the proximity of the basilic vein to the brachial artery and the median nerve, this vein should only be considered if no other vein is more prominent. To emphasize, always survey both arms with a tourniquet if needed for the presence of the median or cephalic veins before choosing the higher risk basilic vein. Most nerve injuries that result from punctures in the antecubital area occur during attempts to puncture the basilic vein. In addition to nerves, the basilic veins' close proximity to the brachial artery subjects the participant to the risk of an arterial nick and subsequent hemorrhage. Should this artery be pierced unknowingly, the consequences can range from a barely perceptible bruising to a severe hemorrhage, which if undetected, can lead to a compression nerve injury. Therefore, when considering punctures to the inside aspect of the antecubital area, phlebotomists should attempt to locate the brachial artery by feeling for a pulse and avoid punctures in the area if the artery lies precariously close to the basilic vein. Although those new to phlebotomy may only feel confident puncturing veins that are visible, this is a luxury not all subjects provide. With time and repetition, confidence in attempting to access veins that are only palpable will increase. However, phlebotomists should never blindly stick for a vein that is neither visible nor palpable. Never puncture anything that has a pulse.

Supplies should always be kept within reach. Should more gauze or tubes be needed during the puncture, reaching for supplies that are not within arm's length puts the participant at risk of injury. Should a tube holder, tourniquet, tube, etc. fall, let the participant see you discard it. Keeping extra supplies within reach prevents this disturbing perception and retains participant confidence. Once a suitable vein in the forearm is identified and a rubber tourniquet is applied, thoroughly cleanse the skin (i.e. insertion site) with a 70% isopropyl alcohol pad. The proper way to cleanse the skin is from the center outward and downward (from the intended needle insertion site, i.e., the most clean area, cleaning outward to the dirty fringes of the site). Be sure to cover the whole area. Ensure the skin area is in contact with the disinfectant for at least 30 seconds and allowed to air dry completely. The drying process kills some bacteria and prevents a burning sensation upon needle insertion. Once the site has been cleansed, do not touch the site. Repalpating for the vein after cleansing contaminates the site and risks infection.

The most common method of drawing blood on prominent veins of the antecubital area in our laboratory is through the use of winged blood collection sets, which are also known as "butterfly" sets. The 23Gx3/4 Vacutainer Brand Blood Collection Set is most common gauge used. Their lightweight design of the butterfly apparatus makes it easy to manipulate, and the wings allow for a lower angle of insertion and greater control than what a syringe or

tube holder can offer. This is especially useful when attempting to access veins of the hand or forearm. Note: If a butterfly set is used in the hand, the tourniquet is usually placed midway up the forearm. Like syringes, winged-collection sets allow the operator to see an immediate flash of blood in the tubing, indicating that the vein has been successfully accessed.

Before using any needle it is important to inspect the paper seal which manufacturers apply to assure sterility. If the seal is broken, its sterility is in question and the needle must be discarded. To prepare the needle for assembly, hold the needle at opposite ends and twist the smaller protective cap off exposing a tight-fitting vinyl sleeve that encases the short “back end” of the needle. Discard the cap. Thread the exposed end into the threads of the tube holder. If the needle is exposed and comes in contact with any surface at any point during the assembly, its sterility is lost and the needle must be discarded. Once a suitable vein has been selected, loosen the tourniquet and assemble the equipment. Leaving the tourniquet on longer than one minute creates hemoconcentration (the static pooling of blood within the veins below venous constriction due to prolonged tourniquet application) and alters the specimen before it is even collected. On participants with prominent veins, it may not be necessary to release the tourniquet as long as vein selection, equipment assembly, site cleansing and venous access can be accomplished within one minute of tourniquet application. However, if it is anticipated that the process may take longer than one minute, release the tourniquet to prevent hemoconcentration. If vein selection alone takes longer than one minute, release the tourniquet and allow two minutes to pass before retightening it and performing the puncture. This allows hemoconcentration to dissipate.

Access the vein by stretching the skin by pulling downward on the arm from below the intended puncture site, but not in such a way that it will obstruct the tube holder. When the skin is taut, the needle passes through it much easier and with significantly less sensation. This technique anchors the vein to prevent it from rolling away from the needle and minimizes the pain of the puncture. With a steady advance and a forward motion, guide the needle, bevel up, into the skin and the vein at an angle of 20 to 30 degrees. Avoid a slow, timid puncture as this will increase participant discomfort. Likewise, don't use a rapid, jabbing motion as this will make passing entirely through the vein likely. Once the needle is anticipated to be within the vein, use the non-dominant hand to maintain the position of the needle by resting the backs of the fingers on the participant's forearm for support. With the dominant hand, attach the syringe to the needle. As you begin to apply suction, use the flared wings of the butterfly needle to counteract the pulling pressure exerted on the bottom of the tube to maintain needle position. Failure to do so may drive the entire needle assembly forward, advancing the tip of the needle through the other side of the vein.

Release the tourniquet once blood flow has been established to prevent hemoconcentration from affecting the results if it is anticipated that the flow will not be interrupted. If blood flow is not established, the needle may be improperly positioned in the vein, or the vein may be too small for the size of the needle or for a vacuum-assisted draw. Strict limitations to needle relocation are necessary to prevent participants from injury. Since excessive needle relocation in the area of the basilic vein can damage nerves and the brachial artery please adhere to the following limitations. If the needle has penetrated too far into the vein, pull it

back a bit. If it has not penetrated far enough, advance it farther into the vein. Rotate the needle half a turn. Try another syringe to ensure the one selected is not defective. Manipulation other than that recommended above is considered probing. Probing is not recommended as it is painful to the participant. In most cases another puncture in a site below the first site, or use of another vein on the other arm, is advisable. If you are unable to obtain a specimen after >2 punctures the process should be discontinued. Repeated failures frustrate the participant, create anxiety and erode the participant's confidence in the skill and professional judgment of the HPRL staff. Further information on these complications and their recommended solutions are provided at the end of this procedure manual (**Appendix 4**).

Tiger top SST tubes and lavender top EDTA tubes are the two most common tubes used to collect blood in our laboratory. In the event that the study protocol requires you to collect both, it is imperative to fill the EDTA tube last because it contains an additive to prevent the specimen from clotting. It is well documented that additive carryover from one tube to the next (EDTA tube to SST tube) drastically alters results. The number of EDTA (5 ml) and SST (8.3 ml) tubes required will vary depending on the study requirements. In our laboratory, approximately 10 ml (2 teaspoons) of venous blood is usually collected at each sampling time point. However, this may vary according to study requirements. Further information on the proper "order of the draw" has been provided at the end of this procedure manual (**Appendix 5**).

If >1 syringe of blood are to be collected, it is critical that the leur end of the syringe is disconnected from the leur end of the butterfly apparatus without displacing the needle. One of the requirements that acceptable specimens must meet is that tubes with additives are filled to the proper level (at least $\frac{3}{4}$ filled). To maintain needle position while disconnecting, maintain constant pressure on the flared wings of the butterfly apparatus with the non-dominant hand. While doing so, use the dominant hand to grasp the leur end of the syringe and rotate slowly in a counterclockwise fashion. If the tube contains an additive (most of the tubes utilized in the HPRL do contain additives), gently invert it 5 to 10 times as soon as possible. Inverting the tubes assures that the specimen mixes with the additive correctly. Once the appropriate amount of blood has been drawn, place a 2x2 gauze sponge just distal to the insertion site. Use the dominant hand to withdraw the needle from the arm and instruct the participant to manually apply pressure to the puncture site. Discard the needle into a red sharps container. Place the evacuated syringe(s) into a biohazard container. Next reassess the participant, who is currently applying pressure to the puncture site. Ensure they continue this process and remain stationary for a minimum of 5 minutes; do not allow them to bend the arm up as a substitute for pressure. At the conclusion of this observation period assess the participant for superficial bleeding and hematoma formation. After assuring that bleeding from both the vein and skin have stopped, place a piece of hypoallergenic transparent surgical tape over the 2x2 gauze sponge to secure it in place. A 1x3 Stat Strip adhesive bandage can also be placed over the needle insertion site to stop any residual bleeding.

In the event that >3 blood samples are to be drawn from the same participant in the same day, it is recommended that an intravenous catheter for blood withdrawal be substituted for the individual needle stick procedure described above. Beyond the Lab Director, no other parties

are permitted to conduct this process. When starting this procedure, the previously mentioned steps are essentially followed. If you use the BD Insyte Autoguard Catheter, you will need to maintain sterile technique. After removing the clear cap over the needle/catheter tip, you will rotate the base of the catheter 360 degrees to the right. The white button on top of the catheter base should serve as a starting and ending point for the rotation. You will then proceed to insert the catheter into the vein after finding, cleansing the site and placing a tourniquet above it. Once you access the vein, you will see a flash in the base of the chamber. Once you see the flash in the chamber, release the tourniquet. Then, you will proceed to advance the catheter into the vein using the tab on the top of the catheter with your index finger. Your middle and thumb should stabilize the base of the catheter. Once the clear plastic is completely inserted into the skin, the white button should be pressed to retract the needle. Pressure should be applied to the vein using one finger while the needle is being retracted. Next, the needle should be discarded and the clear tubing should be inserted and connected to the colored portion of the catheter. Be sure that blood will flow from the site and that the connection is secured tightly before cleaning the area and securing the catheter with tape.

The indwelling venous catheter should next be flushed with 5-10ml of normal saline to ensure the site is patent. Again, the Lab Director is the only party that is allowed to place, flush, or draw from an indwelling catheter. If puffiness is noted after flushing the site, the Lab Director will remove the catheter immediately. The needle went through the vein and the fluid flushed is in the interstitial space causing the skin to swell. An appropriately placed catheter should yield a good blood return and be flushed without difficulty. If difficulty is noted, the connections are not secure, the blood has clotted or the catheter is against the wall of the vein and the Lab Director will need to pull it (the catheter) out slightly. The safety of the participant, the Lab Director's safety, and the integrity of the blood samples are our main priorities. It is important to note that this flushing process should be repeated every 30 minutes during exercise and every 1 hour after exercise for a period of 2hrs following exercise participation. This differs from the clinical setting, in which flushing every 4 hours is typically sufficient. The reason for this difference is the increase in clotting factors (prothrombin, thrombin, fibrin, fibrinogen, etc) during and following exercise; these factors must be taken into account to ensure that the indwelling venous catheter does not clot off and need to be re-inserted, which would expose the participant to undue trauma. There is a possibility that this procedure could affect electrolyte values in the extracellular fluid. However, the likelihood of such an event is minimized when one considers the ratio between the flushing volume (10ml) versus the blood volume (5000ml) and the fact that 5-10 ml of fluid is drawn from the sample line and discarded (i.e. the burn) prior to collection of each blood sample. Blood electrolytes are also not typically analyzed by the HPRL. For these reasons, and because the only viable alternative to the above listed procedure is to allow the indwelling venous catheter to clot off (removing its' utility and exposing the participant to undue trauma), this is the procedure that will be followed in the HPRL.

Blood Processing

A wide variety of specimen collection tubes are available for the multitude of tests that can be performed on participant blood. Most tubes contain an additive to either prevent or

promote clotting of the specimen. These additives are manufactured and added to tubes under very exacting specifications and can significantly alter test results if the tube is not properly filled or handled during and after collection. The primary factor determining which tube is used within the HPRL is whether serum or plasma is required for the respective test. Serum is the liquid portion of the blood after centrifugation of a specimen that is allowed to clot. The tiger top SST tubes ultimately provide serum after centrifugation. Plasma, on the other hand, is the liquid portion of the blood after centrifugation of a specimen in which an anticoagulant has prevented clot formation. The lavender EDTA tubes can provide either whole blood or plasma depending on if the tube is centrifuged. In rare cases, green top sodium heparin tubes and gray top potassium oxalate/sodium fluoride tubes are utilized within the HPRL. The green top tubes are used for plasma chemistry profiles while the gray top tubes are used exclusively for glucose testing.

Not as many routine tests can be performed on plasma as can be performed on serum. Thus at least two SST tubes (tiger top) are usually collected within the HPRL. Glass activates the clotting mechanism. When collection tubes are made of plastic (which include the tubes now used in the HPRL), an additive is required in order to facilitate the clotting process so that serum can be obtained. The tube must be inverted 5-10 times to enhance the clotting process. These clot activators don't necessarily hasten clotting, but facilitate full clotting that renders a fibrin-free specimen after centrifugation. Some clot activator tubes also contain a thixotropic gel barrier to facilitate separation of the serum from the cells during centrifugation. As the force of centrifugation increases, the gel becomes liquid, migrates to the interface of the serum and cells, and re-solidifies to provide a barrier that prevents contact. The thixotropic gels inserted in the tubes by most manufacturers are inert and impart no interference to test results. Tubes with clot activators yield serum used in a multitude of chemistry profiles (serology and immunology tests).

No other anticoagulant preserves cellular integrity or prevents platelet aggregation as well as EDTA (ethylenediaminetetraacetic acid), making it the anticoagulant of choice for whole blood hematology determinations and immunohematology testing (ABO, Rh typing, antibody screens, etc.). EDTA (lavender top) disrupts the clotting process by binding calcium, an essential element for clotting to occur. Lavender-topped EDTA tubes contain dried K₂ (di-potassium) or K₃ (tri-potassium) salts and must be inverted 5-10 times upon filling. These tubes remain well mixed and are not centrifuged prior to testing. Most standard organizations recognize K₂ EDTA as the anticoagulant of choice for blood cell counts. The HPRL currently has a combination of K₂ and K₃ EDTA tubes as the latter are slowly being phased out.

Laboratories that comply with the standards for laboratory testing (i.e., Physicians Preferred) have strict and well-defined specimen rejection criteria. One of the requirements that acceptable specimens must meet is that tubes with additives are filled to the proper level. Tube manufacturers calibrate the amount of additive in each tube to obtain accurate results based on a full draw. Under filling tubes alters the optimal blood: anticoagulant ratio and can significantly affect test results and misrepresent the participant's actual physiology. When a laboratory (i.e., Physicians Preferred) receives a specimen filled below the minimum acceptable volume, the specimen processor or testing personnel is obligated to request another specimen be drawn. Failing to reject compromised specimens risks reporting inaccurate results and initiating a cascade of events that can affect the diagnosis, medication and/or care of the patient. Collectors who submit under filled specimens to the laboratory put the participant at risk when testing is delayed by the inevitable request for recollection. A far greater risk to the participant is when under filled specimens are accepted and tested by unscrupulous testing personnel who fail to uphold minimum-filled standards. Fortunately,

many tests are not dependent upon the tube being completely filled. Tubes without additives have no minimum fill requirements. However the tubes most commonly used with the HPRL do have additives and must be filled completely.

Vacutainer tubes need to be labeled with the participant's name, study, and testing session using a fine tip sharpie marker. Tubes are placed in a test tube rack and placed in the refrigerator. It is best to let the SST tubes clot for 15-20 minutes before centrifuging for 10-15 minutes. After centrifuging the sample in the HPRL the lab technician needs to have gloves on both hands and safety glasses on before proceeding. If the samples are analyzed in house, the blood is transferred to the biochemistry area. Here it is transferred to appropriately labeled micro-centrifuge tubes that are subsequently stored appropriately labeled freezer boxes in the -80°C freezer. Empty blood collection tubes, pipette tips, and all other materials that came in contact with biological materials are discarded into a red biohazard container. If the samples are sent off to an outside laboratory (i.e. Physicians Preferred) they either need to be transferred to Student Medical Services (where an appropriate requisition form must be completed) or appropriately prepared for transport. "Appropriate preparation" includes the following: Triple package the item. For example blood is collected in tubes-1, over that a zip lock bag-2, and over that an outer container-3 that is used to carry the items. Personal vehicles are acceptable for sample transport so long as DOT regulations are followed. Be aware that if this is done in a private vehicle and there is a spill your insurance will not cover the cost of clean-up.

Upon completion of the blood processing procedure, gloves are discarded into the red biohazard can, safety glasses (if worn) are removed and cleaned, and both hands are washed thoroughly. When followed, these procedures will help to minimize your likelihood of occupational exposure to a pathogen. However, it is important that you realize that even with the best practices, health workers may occasionally be accidentally exposed to blood and other body fluids that are potentially infected with HIV, hepatitis virus or other bloodborne pathogens. Occupational exposure may occur through direct contact from splashes into the eyes or mouth, or through injury with a used needle or sharp instrument. Post-exposure prophylaxis (PEP) can help to prevent the transmission of pathogens after a potential exposure. The steps that follow describe the steps for managing exposure to blood or other fluids that are potentially infected with hepatitis B virus (HBV); hepatitis C virus (HCV) or HIV.

Step 1 – Provide immediate first-aid care to the exposure site

Provide immediate first-aid care as follows.

- Wash wounds and skin with soap and water in the provided biosafety sink. Do not use alcohol or strong disinfectants.
- Let the wound bleed freely.
- Do not put on a dressing.
- Flush eyes, the nose, the mouth and mucous membranes with water for at least 10 minutes.

Step 2 – Determine the risk associated with the exposure

Determine the risk associated with the exposure by considering:

- the type of fluid; for example, blood, visibly bloody fluid, other potentially infectious fluid, or tissue and concentration of virus;
- the type of exposure; for example, there is a higher risk associated with percutaneous injury with a large, hollow-bore needle, a deep puncture, visible blood on the device, a needle used in an artery or vein, and exposure to a large volume of blood or semen, and less risk associated with exposure of mucous membranes or nonintact skin, or exposure to a small

volume of blood, semen or a less infectious fluid (e.g. cerebrospinal fluid).

Step 3 – Evaluate the source of the potential exposure

To evaluate the source of the potential exposure:

- assess the risk of infection, using available information;
- test the source person whenever possible, and only with his or her informed consent; but
- do not test discarded needles or syringes for virus contamination.

Step 4 – Report the incident

After the incident, refer the exposed person to the WTAMU (and possibly TAMUS) Biosafety offices. Those offices will designate a trained service provider who can give counseling, evaluate the risk that transmission of bloodborne pathogens has occurred, and decide on the need to prescribe ARV drugs or hepatitis B vaccine to prevent infection. The contact information for those individuals follows.

TAMUS Biosafety Officer
Dr. Bruce Whitney
Email: brucewhitney@tamus.edu
Phone: 806-651-2740

Note: **When in doubt, REPORT!!!** WTAMU considers any unusual event as an incident. The EHS Chemical Hygiene Plan (SOP No. 24.01.01.W1.33AR) provides additional clarification on this issue:

- An incident has neither a positive nor a negative connotation and is by definition an event or occurrence.
- ALL incidents at WTAMU will be investigated. These investigations allow WTAMU students, faculty, and staff the opportunity to participate in the safety culture WTAMU has created, and ensure that any occurrence, no matter how small it may seem, is critically examined to confirm WTAMU is providing safe laboratory conditions.
- Initial Incident Investigations will be conducted with the Laboratory Supervisor and EHS.

If a pattern of unsafe procedures or conditions emerges the following procedures will apply:

- The VP of Research and EHS will meet with the Laboratory Supervisor to discuss concerns as indicated by prior Incident Investigations.
- If further actions are necessary, the VP of Research and EHS will meet with the associated program Department Head and, if necessary, the Dean of the associated College in order to bring resolution to the identified concerns.
- An Incident Report Form appears at the end of this procedure manual (**Appendix 6**).
- A Workers' Compensation First Report of Injury Form appears at the end of this procedure manual (**Appendix 7**).

Both the incident report and the evaluation of the risk of exposure should lead to quality control and evaluation of the safety of working conditions. Take correctional measures to prevent future exposure!

Thank you for reading the HPRL Phlebotomy procedures manual to completion. Please complete the post-test quiz that appears at the end of this procedure manual (**Appendix 10**).

Record Retention

No official state records may be destroyed without permission from the Texas State Library as outlined in [Texas Government Code, Section 441.187](#) and [13 Texas Administrative Code, Title 13, Part 1, Chapter 6, Subchapter A, Rule 6.7](#). The Texas State Library certifies Agency retention schedules as a means of granting permission to destroy official state records.

West Texas A & M University Records Retention Schedule is certified by the Texas State Library and Archives Commission. West Texas A & M University Environmental Health and Safety will follow [Texas A & M University Records Retention Schedule](#) as stated in the Standard Operating Procedure [61.99.01.W0.01 Records Management](#). All official state records (paper, microform, electronic, or any other media) must be retained for the minimum period designated.

Training

West Texas A & M University Environmental Health and Safety will follow the Texas A & M University System Policy [33.05.02 Required Employee Training](#). Staff and faculty whose required training is delinquent more than 60 days will have their access to the Internet terminated until all trainings are completed. Only Blackboard and Single Sign-on will be accessible. Internet access will be restored once training has been completed. Student workers whose required training is delinquent more than 30 days will need to be terminated by their manager through Student Employment.

Related Statutes, Policies, or Requirements

Contact Office

WTAMU Environmental Health and Safety
(806) 651-2270

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The above references were utilized to construct the majority of this resource manual. In addition, the following clinical research laboratories provided us with their current best practices:

1. Texas A&M University, Department of Health & Kinesiology, Exercise & Sport Nutrition Laboratory
2. University of New Mexico, Department of Internal Medicine, Clinical Education Unit
3. Texas Tech University, Health Sciences Center, Clinical Laboratory Sciences Program

*Their assistance on the construction of this SOP is sincerely appreciated.

Appendix 1:

Infection prevention and control practices

Infection prevention and control practices

Do	Do not
DO carry out hand hygiene (use soap and water or alcohol rub), and wash carefully, including wrists and spaces between the fingers for at least 30 seconds (follow WHO's 'My 5 moments for hand hygiene' ^a)	DO NOT forget to clean your hands
DO use one pair of non-sterile gloves per procedure or patient	DO NOT use the same pair of gloves for more than one patient DO NOT wash gloves for reuse
DO use a single-use device for blood sampling and drawing	DO NOT use a syringe, needle or lancet for more than one patient
DO disinfect the skin at the venepuncture site	DO NOT touch the puncture site after disinfecting it
DO discard the used device (a needle and syringe is a single unit) immediately into a robust sharps container	DO NOT leave an unprotected needle lying outside the sharps container
Where recapping of a needle is unavoidable, DO use the one-hand scoop technique (see Annex G)	DO NOT recap a needle using both hands
DO seal the sharps container with a tamper-proof lid	DO NOT overfill or decant a sharps container
DO place laboratory sample tubes in a sturdy rack before injecting into the rubber stopper	DO NOT inject into a laboratory tube while holding it with the other hand
DO immediately report any incident or accident linked to a needle or sharp injury, and seek assistance; start PEP as soon as possible, following protocols	DO NOT delay PEP after exposure to potentially contaminated material; beyond 72 hours, PEP is NOT effective

PEP, post-exposure prophylaxis; WHO, World Health Organization.

^a <http://www.who.int/gpsc/5may/background/5moments/en/index.html>

Appendix 2:

Explaining the procedure to a patient

Introduction:

Hello, I am _____ I work at this health-care facility. I am trained to take blood for laboratory tests (or medical reasons) and I have experience in taking blood. I will introduce a small needle into your vein and gently draw some blood for _____ tests. (Tell the patient the specific tests to be drawn). Do you have any questions? Did you understand what I explained to you? Are you willing to be tested?

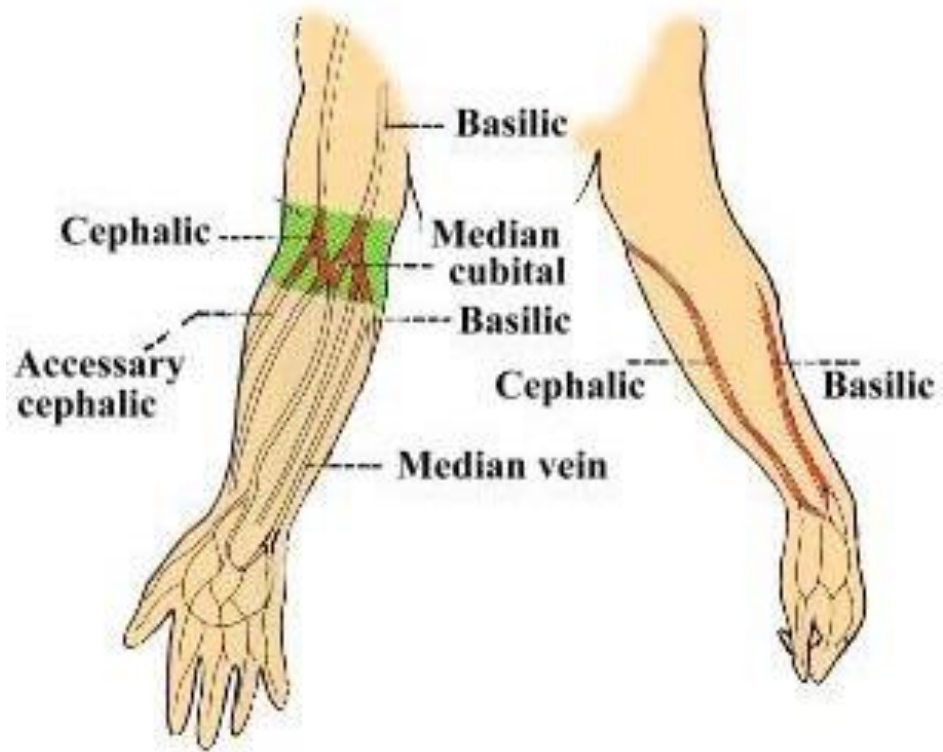
Please sit down and make yourself comfortable. Now, I will ask you a few questions so that both of us feel comfortable about the procedure.

- Have you ever had blood taken before?
 - (If yes) How did it feel? How long ago was that?
- Are you scared of needles?
- Are you allergic to anything? (Ask specifically about latex, povidone iodine, tape.)
- Have you ever fainted when your blood was drawn?
- Have you eaten or drunk anything in the past two hours?
- How are you feeling at the moment?

Shall we start? If you feel unwell or uncomfortable, please let me know at once.

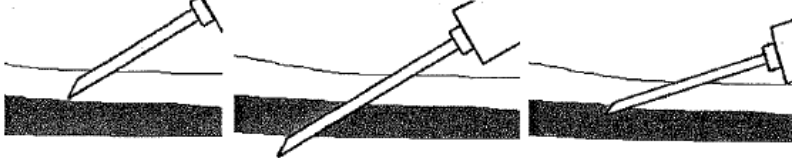
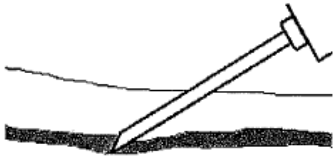

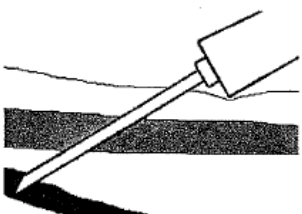
Appendix 3:

Potential Venipuncture Sites



Appendix 4:

Common Complication & Potential Problems

Common Complications & Potential Problems	Suggested Action
 <p><i>Depth or angle of needle is resulting in no blood or very little blood being drawn.</i></p>	<p>Change the position of the needle.</p> <ol style="list-style-type: none"> 1. Move it forward (it may not be in the lumen), OR 2. Move it backward (it may have penetrated too far), OR 3. Adjust the angle (the bevel may be up against the vein wall)
<p><i>A tourniquet that is too tight may obstruct the flow of blood.</i></p>	<p>Loosen the tourniquet.</p>
<p><i>A defective tube with no vacuum will not draw blood effectively.</i></p>	<p>Try another tube.</p>
<p><i>Veins may roll away from point of the needle and puncture site.</i></p>	<p>Anchor the vein below the site during venipuncture.</p>
 <p><i>Collapsed vein will lose pressure and blood flow will cease.</i></p>	<p>Re-secure the tourniquet to increase venous filling. If this is not successful, remove the needle, take care of the puncture site, and redraw.</p>
 <p><i>Hematoma forms under the skin.</i></p>	<p>Release the tourniquet immediately and withdraw the needle. Apply firm pressure. Choose another site.</p>
 <p><i>Blood is bright red (arterial) rather than venous.</i></p>	<p>Apply firm pressure for more than five minutes. Choose another site.</p>

Appendix 4 (continued):

Summary of risks and risk-reduction strategies

Risk	Type of risk	Risk reduction strategy
Patient/ blood donors	Exposure to bloodborne viruses through reuse of needles, syringes and lancets, contaminated work surfaces	<ul style="list-style-type: none"> • Hepatitis B vaccine for workers • Sterile single-use devices only • Safety-engineered devices • Clean work surfaces with disinfectant
	Infection at blood sampling site	<ul style="list-style-type: none"> • Perform hand hygiene • Clean patient's skin with 70% Isopropyl alcohol and allow to dry • Use sterile needle and syringe removed from the packaging just before use
	Pain at blood sampling site	<ul style="list-style-type: none"> • Well-trained person should take the blood sample • Venepuncture is less painful than heel-pricks in neonates • Use needle of smaller gauge than the selected vein
	Haematoma or thrombus	<ul style="list-style-type: none"> • Enter vessel at an angle of 30 degrees or less • Use gauge of needle smaller than the vein • Apply pressure to a straight arm for 3–5 minutes after drawing blood
	Extensive bleeding	<ul style="list-style-type: none"> • Take a history to identify patients on anticoagulants and with a history of bleeding • Use a gauge of needle smaller than the vein
	Nerve damage (8, 10)	<ul style="list-style-type: none"> • Avoid finger-pricks for children • Use antecubital vessels when possible • Avoid probing
	Vasovagal reaction Syncope, fainting (8, 10)	<ul style="list-style-type: none"> • Hydrate patient, take postural blood pressure if dehydrated • Reduce anxiety • Have patient lie down if the person expresses concern • Provide audio-visusal distraction
	Allergies	<ul style="list-style-type: none"> • Ask about allergies to latex, iodine and alcohol before starting the procedure
Health worker	Needle or sharps injury during or after the procedure Breakage of blood containers Splashes (rare)	<ul style="list-style-type: none"> • Use safety devices such as needle covers, tube holders that release needles with one hand and safety lancets • Avoid two-handed recapping and disassembly • Place sharps container in sight and within arm's reach • Dispose of used sharps immediately
	Exposure to blood	<ul style="list-style-type: none"> • Hepatitis B vaccination • Wear gloves • Use evacuated tubes and transfer devices when drawing multiple tubes • Follow protocol for exposure to body fluids and report incident, even if post-exposure prophylaxis is not desired • Cover broken skin area with a waterproof dressing

Appendix 5:

Recommended order of draw for plastic vacuum tubes

Recommended order of draw for plastic vacuum tubes

Order of use ^a	Type of tube/usual colour ^b	Additive ^c	Mode of action	Uses
1	Blood culture bottle (yellow-black striped tubes)	Broth mixture	Preserves viability of microorganisms	Microbiology – aerobes, anaerobes, fungi
2	Non-additive tube			
3	Coagulation tube ^d (light blue top)	Sodium citrate	Forms calcium salts to remove calcium	Coagulation tests (protime and prothrombin time), requires full draw
4	Clot activator (red top)	Clot activator	Blood clots, and the serum is separated by centrifugation	Chemistries, immunology and serology, blood bank (cross-match)
5	Serum separator tube (red-grey tiger top or gold)	None	Contains a gel at the bottom to separate blood from serum on centrifugation	Chemistries, immunology and serology
6	Sodium heparin (dark green top)	Sodium heparin or lithium heparin	Inactivates thrombin and thromboplastin	For lithium level use sodium heparin, for ammonia level use either
7	PST (light green top)	Lithium heparin anticoagulant and a gel separator	Anticoagulants with lithium, separates plasma with PST gel at bottom of tube	Chemistries
8	EDTA (purple top)	EDTA	Forms calcium salts to remove calcium	Haematology, Blood Bank (cross-match) requires full draw
9	Blood tube (pale yellow top)	Acid-citrate-dextrose (ACD, ACDA or ACDB)	Complement inactivation	HLA tissue typing, paternity testing, DNA studies
10	Oxalate/fluoride (light grey top)	Sodium fluoride and potassium oxalate	Antiglycolytic agent preserves glucose up to five days	Glucoses, requires full draw (may cause haemolysis if short draw)

ACD, acid-citrate-dextrose; DNA, deoxyribonucleic acid; EDTA, ethylenediaminetetraacetic acid; HLA, human leucocyte antigen; PST, plasma separating tube.

^a "1" indicates draw first, and "10" draw last (if used).

^b Verify with local laboratory in case local colour codes differ.

^c Gently invert tubes with additives to mix thoroughly; erroneous test results may be obtained when the blood is not thoroughly mixed with the additive.

^d If a routine coagulation assay is the only test ordered, then a single light blue top tube may be drawn. If there is a concern about contamination by tissue fluids or thromboplastins, then a non-additive tube can be drawn before the additive tube. The PST tube contains lithium heparin anticoagulant and a gel separator; if used, draw in the order shown.

Source: Table adapted with permission from WebPath, Mercer University, United States (<http://library.med.utah.edu/WebPath/webpath.html>). Order is based on United States National Committee for Clinical Laboratory Standards consensus (43).

Equipment Involved / ID Number

Describe Possible Causes

Equipment

Tools / PPE

Environment

Procedure

Personnel

Other

Causal Factors

Recommendations

Corrective Actions/Preventive Actions

Person Responsible

Due Date

Corrective Actions/Preventive Actions	Person Responsible	Due Date
<hr/>	<hr/>	<hr/>
<hr/>	<hr/>	<hr/>
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<hr/>	<hr/>	<hr/>

Supervisor Signature

Date

Investigator Signature

Date

Supervisor Comments

APPENDIX 7:

Workers' Compensation

First Report of Injury

Name _____ UIN/Buf ID _____.

Address _____ Phone _____.

Full Time _____ Part Time _____ Student _____

Date of injury _____ Time injury occurred _____

First day employee had lost time _____

Date supervisor or employer was first notified of injury _____.

Where did the accident occur: Premises _____ State _____.

City _____ County _____ Zip _____

Did the accident occur on employer's premises Yes _____ No _____

Did the accident occur at regular place of employment Yes _____ No _____

Describe fully how the accident occurred; state what the employee was doing when injured

Was accident caused by failure to use or observe safety appliance or regulation Yes _____ No _____.

a. Name the safety appliance or regulation provided _____.

b. Was it in use at the time Yes _____ No _____

If injury was caused by machine, tool, etc., give name thereof _____

a. Kind of power used to operate _____.

b. Part on which accident occurred _____.

Names and addresses of witnesses

Describe injury or illness in detail _____

Part of body injured or exposed _____

Name and address of physician _____

Name and address of hospital _____

Expected length of disability _____

Has injured returned to work Yes _____ No _____ If yes, when _____

Any change in wage or occupation due to injury Yes_____No_____

Employee

Date

Department

Completed by

Date

WORKERS' COMPENSATION
Witness Report

Claim No. _____
Employee _____
Employer _____
Date of Injury _____

Name _____ Telephone _____

Address _____

Employer _____ Employer Phone _____

On _____, 20____, at _____ a.m./p.m., I was at (state clearly your location)

_____.
when an accident involving the above employee is alleged to have occurred.

(check one)

I saw the accident and it occurred in the following manner: _____

I did not see the accident, but information given to me by _____
_____ (injured employee or witness) indicates it occurred as follows:

I have no knowledge of the alleged incident

Signature,

Date

Appendix 8:

Phlebotomy Training

The Lab Director considers the competency of all Research Assistants who draw blood in the WTAMU HPRL to be a top priority. Their competence is verified by a multi-step process. The first step includes self-paced review of the written information that is provided on pages 3 – 16 of this procedure manual. The second step is completion of the written mastery quiz, which appears on pages 17 -18 of this procedure manual. The third step is practical skills training/assessment, which are conducted on a phlebotomy training arms. The fourth is a practical skills verification (provided by PI, who is required to have completed phlebotomy training in a clinical laboratory or from a nationally recognized certificate granting program and to required to provide documentation of said training to both the Department Head and Dean of his/her academic unit). This verification requires Research Assistants to complete a minimum of three successful venipunctures (on the training arm) under the PI's supervision. The fifth and final practical skills verification will take place on an actual research subject, who has completed the appropriate Informed Consent document and reported to the HPRL for research data collection.

By definition, “Research Assistants” must be university students. They may/may not be university employees. As such, non-employees must be aware that they are not covered by the university's liability insurance and are operating at the own discretion.

Upon verification of these five steps, Research Assistants are considered to be competent in phlebotomy and are cleared to draw on research participants that have completed the Informed Consent document. In that instance, Research Assistants are still advised that they alone are responsible for knowing their own capabilities and maintaining aseptic techniques. Should they encounter a research participant who presents with vessels or otherwise that make them uncomfortable, it is their responsibility to seek the assistance of qualified medical personnel (housed downstairs @ Student Medical Services) or from the PI.

In addition to this training, all lab personnel are also required to complete CPR/AED/First Aid Certification, BBP (Bloodborne Pathogen) Training, BL2 (Biosafety Laboratory Level 2) Training, CITI (Collaborative Institutional Training Initiative) Training.

West Texas A & M University Environmental Health and Safety will follow the Texas A & M University System Policy [33.05.02 Required Employee Training](#). Staff and faculty whose required training is delinquent more than 90 days will have their access to the Internet terminated until all trainings are completed. Only Blackboard and Single Sign-on will be accessible. Internet access will be restored once training has been completed. Student workers whose required training is delinquent more than 90 days will need to be terminated by their manager through Student Employment.

Appendix 9:

Immunization Status & Liability Insurance Requirements

Blood samples need to be collected by an experienced nurse/phlebotomist/research assistant who has received blood borne pathogen training using standard phlebotomy procedures and has been vaccinated for Hepatitis B. Note: The hepatitis B vaccine was added to the annual childhood immunization schedule endorsed by the Centers for Disease Control and Prevention (CDC), American Academy of Pediatrics (AAP), and American Academy of Family Physicians (AAFP) in 1995. Individuals born on (or after) this date have likely already received the HBV vaccine. Individuals who were born prior to this date or are unsure about their HBV immunization status for any reason are advised to contact the West Texas A&M University Environmental Health and Safety office (EHS) at (806)651-2270. In the event it is determined a nurse/phlebotomist/research assistant has not been previously immunized the HBV immunization (provided as a series of 3 shots over a 6 month period) will be made available through Student Medical Services (SMS). If there is any doubt as to the nurse/phlebotomist/research assistant's immunization status, they will be assumed to be non-vaccinated and required to receive the HBV immunization (as described above).

In addition each person needs to go through phlebotomy training, which is offered by the Lab Director on an as-needed basis and must be repeated by all research assistants annually. Original documentation of said training will reside in the lab; copies of said training will be furnished to both the SES Department Head and to EHS.

All parties must also have evidence of liability insurance that covers phlebotomy and other lab procedures. Faculty, instructors, and lab assistants that are paid employees are already covered under worker's compensation insurance within the course and scope of their employment duties and the limits of Texas tort liability. Enrolled students (both undergraduate and graduate) require supplemental insurance. This insurance has been made available to the HPRL through the office of Business & Finance – Risk Management. The current cost (2013) is \$11/lab assistant/ calendar year. This fee is set up as a specific course fee and billed to a local account number, allowing the insurance premium to be applied directly to a student's bill. The enrollment date is also open, meaning new student phlebotomists can enroll at any time. Specific information on the limits of this liability (which apply to faculty, instructors, lab assistants, and student phlebotomists) follow:

Limits of Liability: The limit of liability per state entity and individual as found in *Civil Practice and Remedies Code Title 5 Government Liability Chapter 101* and *Civil Practice and Remedies Code Chapter 104 State Liability For Conduct Of Public Servants* as applicable to "each claim" is the limit of the self-retained medical malpractice liability for all damages because of each claim or suit against each TAMUS Member Entity and/or employee covered by the Plan.

<u>Individual Employee</u>	<u>Limits</u>
Each Claimant	\$100,000
Each Single Occurrence	\$300,000

<u>State Entity</u>	<u>Limits</u>
Each claimant	\$250,000
Each Single Occurrence	\$500,000

Appendix 10:

Phlebotomy self-study module: Post-test Mastery quiz

1. When performing venipuncture in adults, the technician should expect to use a 21 gauge needle
 - a. True
 - b. False
2. When inserting a needle into a vein, the bevel should be facing downwards (toward the skin).
 - a. True
 - b. False
3. When drawing blood, the tourniquet should be released...
 - a. As soon as venipuncture takes place
 - b. When blood begins to flow into the last tube
 - c. After two minutes
 - d. After the needle is removed
4. The vein of choice for drawing blood in most adults is the:
 - a. Brachial vein
 - b. Median cubital vein
 - c. Basilica vein
 - d. Femoral vein
5. In which of the following situations is hemolysis likely to occur?
 - a. Vigorous shaking of drawn tubes of blood
 - b. Drawing through a 25-gauge needle into a syringe and then directly into an evacuator tube
 - c. Pulling back the syringe plunger too forcefully
 - d. All of the above
6. Which statement about arteries and veins is FALSE?
 - a. Veins are located more superficially than arteries
 - b. Arteries do not have valves, whereas veins do
 - c. The walls of veins are thicker than the walls of arteries
 - d. Blood found in veins is darker than blood found in arteries
7. Which of the following draws was done according to the recommended "order of the draw"?
 - a. lavender, green, red
 - b. green, light blue, lavender
 - c. red, green, lavender
 - d. red, lavender, light blue
8. What is the primary reason for applying heat to an intended puncture site?
 - a. It reduces the discomfort of the puncture
 - b. It helps distend the vein, making it easier to see
 - c. It reduces the risk of hematomas developing
 - d. It helps stabilize the vein and prevents it from rolling.

9. Why is it important for the phlebotomist to palpate a vein?
 - a. It helps to determine the depth of the vein
 - b. It helps to determine the direction of the vein
 - c. It helps to determine what size of needle to select
 - d. All of the above

10. What is the most appropriate angle of entry for most venipunctures?
 - a. 10 degrees
 - b. 20 degrees
 - c. 30 degrees
 - d. 60 degrees

11. Coercion of a patient to allow blood to be drawn may be considered assault.
 - a. True
 - b. False

12. Which of the following veins is not located in the arm?
 - a. Basilica
 - b. Cephalic
 - c. Saphenous
 - d. Median cubital

13. What is the most likely cause of a burning sensation felt by a patient during venipuncture?
 - a. The needle hit a nerve
 - b. The needle struck a valve within the vein
 - c. The tourniquet was applied too tightly
 - d. The cleansing agent was not allowed to sufficiently dry