PhenX Toolkit

Data Collection Worksheet

Please Note: The Data Collection Worksheet (DCW) is a tool to aid integration of a PhenX protocol into a study. The PhenX DCW is not designed to be a data collection instrument. Investigators will need to decide the best way to collect data for the PhenX protocol in their study. Variables captured in the DCW, along with variable names and unique PhenX variable identifiers, are included in the PhenX Data Dictionary (DD) files.

Factor VIII Activity in Plasma Using the Chromogenic Substrate Assay

Sample Collection

The working group (WG) recommends that investigators follow the sample collection procedures outlined in Lippi et al. (2012) to ensure quality specimens for coagulation testing. These recommendations include basic criteria for venipuncture (e.g., proper patient identification, use of correct techniques, appropriate devices and needles) as well as additional guidance for critical parameters which can affect the outcome of clot-based tests. These critical parameters include prevention of prolonged venous stasis, collection of nonhemolyzed samples, order of blood draw, and appropriate filling and mixing of collection tubes.

Additionally, the WG highlights that blood should be collected by direct venipuncture into 3.2% sodium citrate tubes and filled within 11% of fill line. A second tube should be collected. A discard tube should be drawn if using a winged butterfly collection system.

Sample Processing

The WG recommends that investigators follow the sample collection procedures outlined in Adcock Funk et al. (2012). The procedures include that:

- unprocessed or processed sodium citrate samples remain capped and at room temperature until testing,
- samples should not be refrigerated or stored on ice or in an ice bath,
- samples should be transported vertically, and
- samples should not be agitated during transportation to avoid remixing of components.

Additionally, samples can be transported and stored as:

- unprocessed sodium citrate whole blood samples,

- whole blood samples centrifuged and maintained in sodium citrate tubes, or
- plasma processed by centrifugation and aliquoting into a second tube.

Ideally, whole blood samples should be processed to platelet poor plasma within 1 hour of collection and assayed within 4 hours of collection.

If centrifuging samples, the centrifuge should be validated so that process results in less than 10,000 platelets/microliter. Centrifuged and processed plasma can be stored at -20^{o} C for 2 weeks and should be transferred to <= -70^{o} C for longer storage, including shipment.

Chromogenic Substrate Assay

The WG notes that there are a number of different assays and instruments that are appropriate to perform the chromogenic substrate assay. Once an assay is chosen for a particular study, the WG recommends that no changes in the protocol be made over the course of the study. Because results can vary with the instrumentation and reagents, the WG recommends that the investigator record the make and manufacturer of equipment, the repeatability and coefficients of variation for the assay, and the reagents used.

Interpretation of Results

The International Society on Thrombosis and Haemostasis (Blanchette et al., 2014) provides the following consensus definitions for the severity of hemophilia A based on plasma levels of Factor VIII activity:

- severe hemophilia A if < 1% of normal,
- moderate hemophilia A if > 1% and < 5% of normal,
- mild hemophilia A if > 5% of normal.

Protocol source: https://www.phenxtoolkit.org/protocols/view/910301