

Blood Collection and Handling

Tube Additives, Tube Type

Most laboratory tests are performed on plasma, serum, or whole blood. To preserve the specimen in the form required by the test, collection tubes contain additives that either prevent coagulation (for plasma and whole blood recovery), or activate coagulation (for serum recovery). Please refer to individual test requirements.

Drawing Order

When multiple tubes are drawn, it is important to prioritize the drawing order to prevent a tube additive from contaminating the next tube and altering the chemical composition of the following specimen. Coagulation tests are highly susceptible to interference from contamination from tissue fluid and tube additives; therefore these tests are usually collected first when a series of tubes are collected. Prior to collecting tests for coagulation (i.e. Blue top tube) a plain **Clear** top tube containing **no additive** must be partially filled and discarded. This “waste” tube prevents tissue thromboplastins from contaminating the Blue top tube. Blue top tubes must be allowed to fill to the line indicated on the tube, exhausting the vacuum. See *Table 1, “Vacutainer Order of Draw”* for proper collection order of vacutainer tubes.

Certain blood collection techniques have been identified as possible sources of error in laboratory testing. Avoid the following sources of test error when collecting blood:

- Tourniquet left on arm for over a minute before blood collection.
- Techniques causing increased trauma to vein or surrounding tissues.
- Drawing from a site below an intravenous port.
- Drawing from a site that is still wet from the antiseptic used to clean the site.
- Inappropriate, expired, or partially filled collection tube.
- Drawing multiple tubes out of order.
- Patient incorrectly identified.
- Tube incorrectly labeled.
- Patient preparation instructions were not followed.

Table 1. Vacutainer Order Of Draw

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| 1. Navy Blue (metals testing) |
| 2. Blood culture bottles / SPS tubes |
| 3. Coagulation tests: <ul style="list-style-type: none">a. Clear top “waste” tubeb. Light Blue top |
| 4. Gold top |
| 5. Plain Red top |
| 6. Dark Green top (heparin) |
| 7. Light Green top |
| 8. Lavender or Pink top |
| 9. Grey top |
| 10. Yellow top (ACD) |

Color top	Code-Volume	Contains	Common Uses	Special Instructions
Light Green	PST - 4.5 ml.	Lithium Heparin, separator gel	Plasma Separator Tube used for many routine chemistry tests.	Immediately after collection, invert the tube 8-10 times. Centrifuge for 10-15 minutes. Using plasma instead of serum speeds the testing process by eliminating fibrin clots and yielding greater specimen volume. Separator gel keeps plasma away from cells.
Green	LIT - 4 ml.	Lithium Heparin	Troponins and other special chemistry tests.	Immediately after collection, invert the tube 8-10 times to mix anticoagulant.
Green	NHP - 6 ml.	Sodium Heparin	Special tests requiring heparinized plasma or whole blood	Immediately after collection, invert the tube 8-10 times to mix anticoagulant.
Lavender	LAV - 4 ml.	K ₂ EDTA	Routine hematology procedures	Immediately after collection, invert the tube 8-10 times to mix anticoagulant.
Pink	7LV - 6 ml.	K ₂ EDTA	Blood Bank / Transfusion testing	Immediately after collection, invert the tube 8-10 times. This tube has a special label for filling in information required for blood bank testing.
Light Blue	BLU - 2.7 ml	Sodium Citrate	Coagulation studies. The inner tube reduces the volume needed to achieve proper blood to anticoagulant ratio.	<ol style="list-style-type: none"> 1. Draw a waste tube before filling this tube to eliminate tissue contamination. 2. The ratio of blood to anticoagulant will affect test, so fill the tube to the top of the label. 3. Immediately invert 8 to 10 times to mix blood and anticoagulant, preventing clot activation. Specimens exhibiting clotting cannot be tested. 4. Centrifuge for 15 minutes at 2800 rpm within 30 minutes of draw. (Centrifuging not necessary for Protime/INR test (PT) if refrigerated & tested within 24 hrs.) 6. Do not disturb "buffy coat" when removing plasma from cells. Plasma must be platelet-poor to prevent test interference from platelet thromboplastins.
Red	RED 7 ml.	Clot activators	Chemistry tests requiring no additives	Mix 8-10 times and allow blood to clot for 30-60 minutes at room temperature before centrifugation. Remove serum from cells promptly after centrifugation.
Royal Blue lilac label	NVE 7 ml for plasma	Na ₂ EDTA. Free of trace metals	Trace element analysis requiring whole blood	If drawing multiple tubes, draw this tube first to avoid contamination from stoppers. Immediately after collection, invert the tube 8-10 times to mix. Send to lab in original tube.
Royal Blue red label	NAV 7 ml for serum	No additive. Free of trace metals.	Trace element analysis requiring serum	Allow to clot for 30 minutes before centrifugation. Remove serum from cells promptly after centrifugation and store in metal-free vial.
Gold	SST 5 ml Serum	clot activator, gel separator	Various tests	Mix 8-10 times and allow blood to clot for 15-30 minutes before centrifugation.
Yellow	ACD 10 ml. Y2B 6 ml.	Solution A Solution B	Whole blood	Immediately after collection, invert the tube 8-10 times to mix. Choose the correct solution for the test required, as tubes look alike.
Other	<i>For information regarding test collection not mentioned here, please contact Client Services @ 331-221-4422</i>			

Whole Blood, Plasma, and Serum Processing

Plasma

- **Draw** a sufficient amount of blood with the required anticoagulant to yield the plasma volume required by the test.
- **Mix.** Immediately after collection, gently mix the blood with the tube additive by inverting 8-10 times. Avoid hemolysis of the specimen during collection and mixing.
- **Place** the specimen in a rack at room temperature and centrifuge within 2 hours of collection. Do not refrigerate the sample until the plasma is separated from the cells.

Platelet-Poor Plasma (Double-spun plasma)

- Draw a Blue-top tube in the proper tube order. Blue top tubes must be filled to the top of the label mark in order to achieve the proper blood to anticoagulant ratio. Mix well by inverting 8-10 times.
- Centrifuge promptly for 15 minutes. Using a plastic pipette, transfer the plasma from the blue top(s) to one or more plastic aliquot tubes, taking care not to disturb the platelet layer that lies on top of the red blood cell layer. Leave a small amount of plasma in the collection tube to be sure you do not pipette out any platelets with the plasma sample.
- Cap and centrifuge the transferred plasma sample for another 15 minutes. While the plasma is spinning again, prepare another labeled plastic aliquot tube for the final platelet-poor plasma sample. Indicate “platelet-poor” or “double-spun” plasma on the label.
- When the second spin is complete, transfer the top 90% of the plasma from the first aliquot tube into the second aliquot tube, taking care not to disturb any platelets that remain in the bottom of the first tube. Discard this first tube, and promptly freeze the platelet-poor plasma that you have prepared.

Serum

- **Draw** a sufficient amount of blood to yield the serum volume required by the test.
- **Mix.** Immediately after collection, mix SST tubes by inverting 8-10 times. This is a very important step! In the past, the glass sides of the collection tubes activated clotting. The switch to plastic tubes due to safety concerns contains a tube additive to activate clotting. For clot formation to occur, tubes must be mixed well. Avoid hemolysis of the specimen during collection and mixing.
- **Clot.** Allow blood to clot by placing in a rack at room temperature for at least 15-30 minutes. Centrifuging specimens before coagulation is complete causes fibrin clots to form in the serum.
- **Centrifuge** within 2 hours of collection. Do not refrigerate the sample until the serum is separated from the cells.

Whole Blood

- **Draw** a sufficient amount of blood with the required anticoagulant tube. To achieve an optimum ratio of blood to anticoagulant, the volume of blood should fill the tube to the line indicated on the vacutainer label.
- **Mix.** Immediately after collection, before clotting can occur, gently mix the blood collection tube by inverting 8-10 times.
- **Store** the sample according to the specific test requirements.