**Lab 1 Isolation of serum and Plasma**

**Serology and Vaccines**

**Serology** is the scientific study of plasma serum and other bodily fluids. In practice, the term usually refers to the diagnostic identification of antibodies in the serum. Such antibodies are typically formed in response to an infection, or antibodies against other foreign proteins, or to one's own proteins.

Serological tests may be performed for diagnostic purposes when an infection is suspected, in rheumatic illnesses, and in many other situations, such as checking an individual's blood type.

There are several serology techniques that can be used depending on the antibodies being studied. These include: ELISA, agglutination, precipitation, complement-fixation, and fluorescent antibodies.

Some serological tests are not limited to blood serum, but can also be performed on other bodily fluids such as semen and saliva, which have (roughly) similar properties to serum.

**Preparation of blood plasma and serum:**

**Blood plasma** is the liquid component of blood, in which the blood cells are suspended. It makes up about 60% of total blood volume. It is composed of mostly water (90% by volume), and contains dissolved proteins, glucose, clotting factors, mineral ions, hormones and carbon dioxide (plasma being the main medium for excretory product transportation). Plasma is the supernatant fluid obtained when anti-coagulated blood has been centrifuged. The blood is mixed with an appropriate amount of anticoagulant like heparin, oxalate or ethylene diamine tetra acetic acid (EDTA). This preparation should be mixed immediately and thoroughly to avoid clotting.

**Blood serum** is blood plasma without fibrinogen or the other clotting factors. Serum is clearer than plasma because of fewer proteins. Proteins are sometimes considered as interfering substances in some tests as they react with the reagent and thereby yield inaccurate results. Serum is the preferred specimen in clinical testing as the interference that may be caused by a plasma specimen because of the presence of an anticoagulant, is eliminated.

**Blood Plasma Preparation**

1. Draw blood into tube(s) containing ~1.8 mg K2EDTA per ml blood (may vary depending on manufacturer). Be sure to draw the full volume to ensure the correct blood-to anticoagulant ratio.

2. Invert vacutainer tubes carefully 10 times to mix blood and anticoagulant and store at room temperature until centrifugation.

3. Samples should undergo centrifugation immediately. This should be carried out for a minimum of 10 minutes at 1000-2000 RCF (generally 1300 RCF) at room temperature

4. This will give three layers: (from top to bottom) plasma, leucocytes (buffy coat), erythrocytes.

5. Carefully aspirate the supernatant (plasma) at room temperature and pool in a centrifuge tube. Take care not to disrupt the cell layer or transfer any cells.

6. Inspect plasma for turbidity. Turbid samples should be centrifuged and aspirated again to remove remaining insoluble matter.

7. Aliquot plasma into vials and store at –80 °C. Ensure that the vials are adequately labeled with the relevant information, including details of additives present in the blood.

**Blood Serum Preparation**

1. Draw whole blood into tube(s) containing no anticoagulant. Draw approximately 2 ½ times the volume needed for use e.g. 10 ml blood for 4 ml serum.

2. Incubate in an upright position at room temperature for 30-45 min (no longer than 60 min) to allow clotting.

3. Centrifuge for 15 min at manufacturer’s recommended speed (usually 1000-2000 RCF). Do not use brake to stop centrifuge.

4. Carefully aspirate the supernatant (serum) at room temperature and pool into a centrifuge tube, taking care not to disturb the cell layer or transfer any cells. Use a clean pipette for each tube.

5. Inspect serum for turbidity. Turbid samples should be centrifuged and aspirated again to remove remaining insoluble matter.

6. Aliquot into vials and store at –80 °C. Ensure that the vials are adequately labeled with the relevant information, including details of additives present in the blood.