EXAMINATION OF BLOOD SMEAR

Clinical significance of blood smear:

1-differential leucocytic count (D.L.C).

2-detection of bacteria in blood in case of bacteremia.

3-detection of blood parasites (babesia, thilleria, anaplasma…..).

4-Detection of shape size and stain of RBCs and WBCs.

5-Detection of inclusion bodies.

6-platlets count.

7-detection of animal species.

Preparation of blood smear:

It is better to make blood smear from freshly drawn blood without anticoagulant. But in case of using blood with anticoagulant you must make blood smear within 15 minutes after adding the anticoagulant to avoid RBCs distortion.

There are two methods for blood film preparation

A) **Slide method** B) **cover glass method**

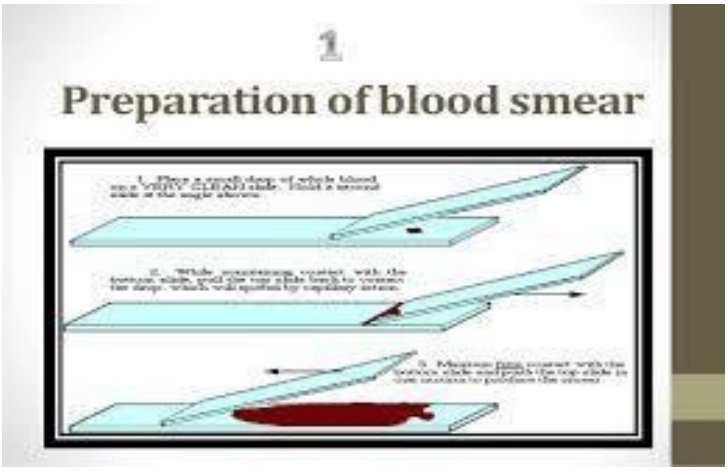
Equipment:

- ☒ Two Clean and dry glass slide
- ☒ Whole blood sample (blood + anticoagulant)
- ☒ Dropper
- ☒ Stain
- ☒ Microscope
- ☒ Filter paper
- ☒ Distilled water

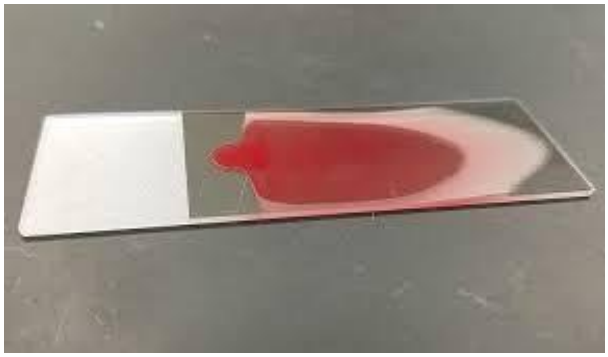
A) Glass slide method:

Procedure:

- Mix the blood sample thoroughly and smoothly in eight figure shape.
- Rest the first glass slide on a flat surface, take a small drop of blood by dropper and put it near one end of the slide.
- Place the end of the other slide (spreader slide) against the surface of the first slide and in front of the blood drop at an angel of 30-45^0.
- Draw the spreader slide gently toward the blood drop till touching it and spread along the 2/3 of the spreader slide edge by the capillary action, then push the spreader slide forward with a steady even motion. The blood will follow making a thin film.
- Let the smear dry slowly by waving in air.



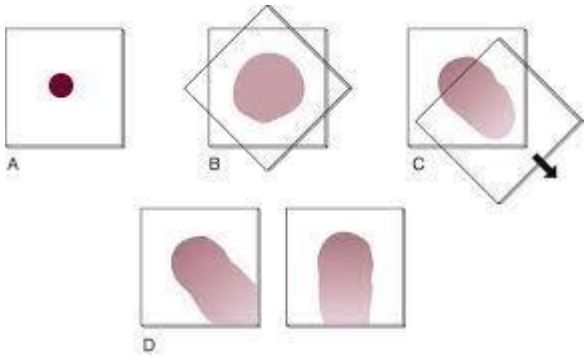
The ideal blood smear composed of three parts head, body and tail.



Ideal blood smear

B) Cover glass method

- Clean two cover glasses and keep in alcohol till time to use and then dry with clean towel and gauze.
- Place a small drop of blood in the center of cover glass.
- Place the second cover glass gently over the first cover glass and the blood will spread between the two cover glasses by capillarity.
- Separate the two cover glasses with a steady parallel motion not upward motion to avoid making holes in the blood smear.
- Allow the smear to dry in air.



N.B.

It is advisable to make two smears at least.

Label each slide using needle or sharp pencil.

Common faults in blood smear

1-too thick smear due to using too large blood drop or wide angel of spread (as in figure C).

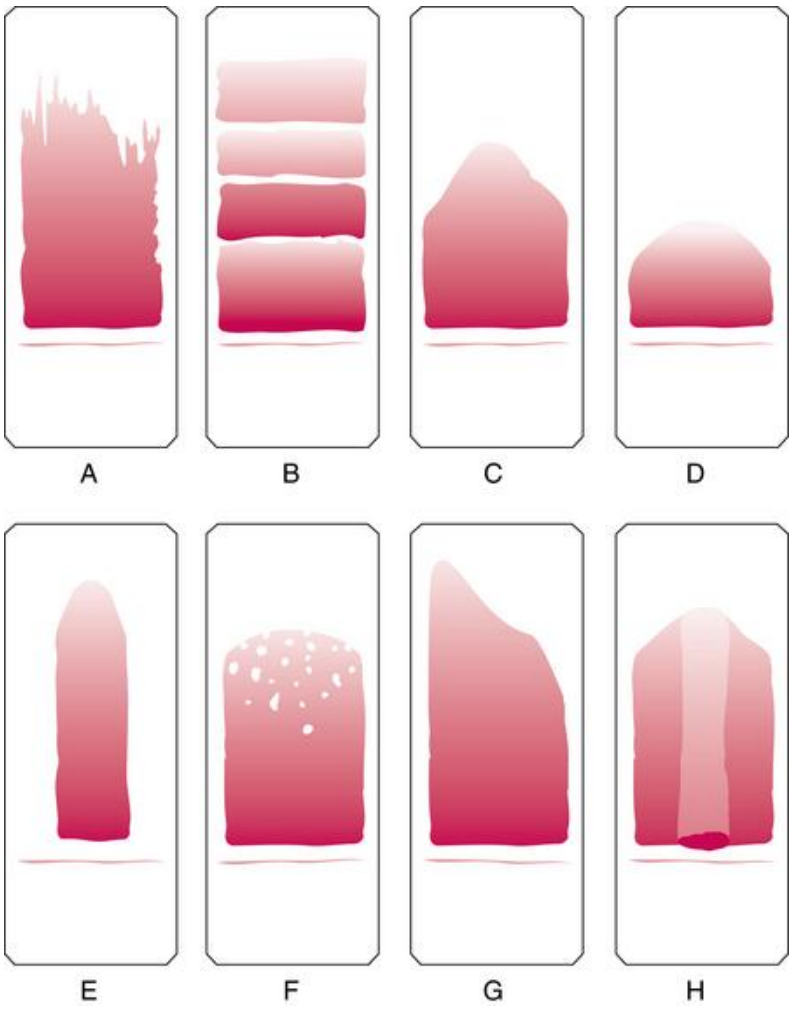
2-too thin smear due to using too small drop of blood or narrow angel of spread (as in figure D).

3-alternative thick and thin bands usually due to spreading with hesitation (as in figure B).

4-streaks throughout the length of the smear due to irregular edge of the spreader slide or presence of dust on the slide (as in figure H).

5-holes free from blood due to presence of grease on slide (as in figure F).

6-very narrow thick slide, smear made before the blood had run along the edge of the spreader slide (as in figure E).



Staining of blood smear:

1. Leishman's stain:

- Cover the smear completely with Leishman's stain and leave it for 1-2 min to fix the smear.
- Add distilled water to the slide twice the volume of the stain already present and gently mix the distilled water and the stain on the slide.
- Allow the mixture to act for 10-15min.
- Wash off the mixture from the slide with distilled water and allow it to remain on the slide for 1 min.
- Pour off the water, dry the smear with filter paper.
- Examine under microscope after dryness with oil immersion objective(100x).

Staining jar

2.Giemsa stain:

- Place the dried blood smear in staining jar containing absolute methyl alcohol for 3-5 min.
- Drain off alcohol and let the slide dry
- Transfer the slide to second jar containing diluted Giemsa stain (1 stain volume/10 distilled water volume) for 15-60 min.
- Wash the slide thoroughly with distilled water.
- Examine under microscope after dryness with oil immersion lens(100x).