

PREPARATION OF THICK MALARIAL BLOOD SMEARS BY THE TAPPING METHOD

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The usual method of preparing thick blood smears for diagnosis and study of malarial parasites consists in using the tip of a toothpick or the point of another slide to run together and mingle three large, adjacent drops of blood, so as to form a round smear about the size of a dime. The parasites are then sought throughout the circular smear because they have not been localized or concentrated in any manner.

In 1954, while the writer was stationed in Monrovia, Liberia with the U. S. Operations Mission to Liberia, the German laboratory supervisor at the Liberian Government Hospital, Miss Freya Weber, showed him the method which was used by her in the preparation of thick blood smears, and which has advantages over the usual method. Her method was as follows:

1. After puncturing the ear lobe or finger, place a very large drop of blood on the slide about one-half inch from one end in the center of the slide.
 2. Tap the opposite end of the slide gently on the table, until the blood flows for about one inch or slightly more along the length of the slide.
 3. Let the slide dry in air, propped up at an angle, so that the distal end of the smear consists of a large blob of dried blood.
 4. Stain as usual with Giemsa stain. (Staining for 60 minutes and washing for 5 minutes are recommended.)
- If any parasites are present, most of them usually flow down into the concentrated

portion and are found comparatively quickly. This method has some disadvantages: (1) the stain does not usually penetrate the concentration completely, although enough of it is stained to permit finding the parasites; (2) it does not readily permit making of more than one thick and thin smear on the same slide; and (3) after the thick smear has dried there is danger of flaking of the crust at the concentrated end, while the slides are being carried around in the field or being jolted around in a jeep, prior to being stained.

While assigned as Malariologist with the U. S. Operations Mission, Kathmandu, Nepal, the writer was confronted with a shortage of slides and with certain personnel problems which necessitated the adoption of the simplest possible method for preparing thick and thin blood smears under the conditions that prevailed. After much experimentation with variations on the tapping method, the following procedure was adopted. (Refer to Figs. A & B.)

1. After puncturing the ear lobe or finger, place one good-sized drop of blood about $\frac{1}{8}$ inch from the long edge, about $\frac{2}{3}$ of the way along the slide.
2. Holding the slide with the long edge parallel to the table top, gently tap the slide vertically against the table top until the blood has flowed transversely across the slide for a distance of $\frac{3}{8}$ to $\frac{3}{4}$ inch.
3. Let the slide dry in air, *flat and horizontally*, so that the blood forms a narrow, even film about $\frac{1}{8}$ inch wide and $\frac{3}{8}$ to $\frac{3}{4}$ inch long.
4. If only the thick smear is desired, no other procedure is necessary; however, if a thin smear is also desired, it should be made *prior* to the thick smear, beginning at about $\frac{3}{4}$ inch from the right end of the slide. It is very important that a *very*

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small drop be used for the thin smear so that only one layer of erythrocytes results.

5. After the thick smear has dried, a second thick (and thin) smear from another child may be made at the other third of the slide.

This latter method has the following advantages: (1) it permits making of both thick and thin smears from two different persons; (2) if thick smears alone are desired, the tapping method avoids the use of a second slide or a toothpick for mingling separate drops of blood; (3) a narrow, uniform blood film is formed which stains uniformly and is easily examined because of its shape; and (4) a saving in slides used is effected due to the doubling up of thick and thin smears.

In practice the slides were kept in boxes of 100 slides each, previously numbered at the upper right with a diamond-pointed pencil. One hundred slides were made consecutively, at the right third of the slide, after which the second hundred smears were made at the left third of the same slides. The smears at the right were given the original numbers of the slides, such as 101, 102, 103, etc., whereas those at the left were prefixed with the letter "L" meaning "left side," such as "L101," "L102," "L103," etc. If desired, "A" can be used instead of "L," it being understood that a letter prefixing the number refers to the smear at the left. No actual number need be written on the slide at the left side.

It is important that the slide be kept flat and horizontal during drying of the thick smear, and in a protected place away from heat of any kind. A cardboard slide tray was found to be very valuable for this purpose; by propping up the two halves of the cover with small sticks or pebbles, drying occurred with a minimum of exposure to flies. A large flour strainer is also useful for keeping flies away from drying slides in the field.

Fixing of thin smears was done as soon as conditions were favorable. This was accomplished by dipping a narrow strip of

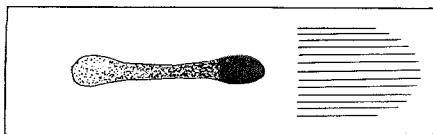


Fig. A

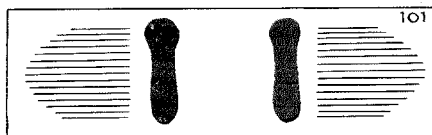


Fig. B

Figure A shows the thick smear prepared by the first tapping method described, with a thin smear at right.

Figure B shows two sets of thick and thin smears prepared as described; the thick smears shaped as usually occurs after tapping.

paper towelling or toilet tissue into methyl alcohol and sliding it rapidly off the thin smear while the slide was held flat in the hand. Slides which are being fixed should be kept away from drafts, windows, and operating fans, as it was found that the alcohol vapor could be wafted over the thick smear, thus fixing and ruining the latter. Once a thick smear has been accidentally fixed with methyl alcohol liquid or vapor, there is no way the smear can be "unfixed" or restored to its original condition, any more than a boiled egg can be restored to its original condition.

SUMMARY. A thick malarial blood smear can be easily prepared by tapping the slide against a table top or other surface, so that a large drop of blood flows transversely across the slide for a distance of $\frac{5}{8}$ to $\frac{3}{4}$ inch, after which the slide is permitted to dry in air in a flat position. A narrow, even film results which is very convenient in searching for malarial parasites. The need for mingling several adjacent drops of blood with some object, as usually done, is therefore obviated.