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AN IMPROVED TECHNIQUE FOR SECTIONING HAIRS*

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While most of the morphological features of hair, both animal and human, can be distinguished by an experienced microscopist without preparation of either transverse or longitudinal sections, it is some, times necessary and usually desirable to observe the hair at least in cross section. This becomes especially true when dealing with animal hairs, the transverse aspect of which is often highly diagnostic of a family with modifications in the genera. Many techniques have been described for this operation, but in practice much difficulty is encountered in embedding the hair so that it gives a true cross section which is representative. Embedding in paraffin¹ alone ordinarily allows the hair to bend ahead of the cutting edge, leading to an angular section which, when observed transforms a circular form into an apparent elliptical shape thereby distorting the true contour of the fiber. Embedding in collodion² is somewhat more successful, but the collodion coated hair must ordinarily be mounted secondarily in paraffin in order to make it rigid. One is thereby confronted with the same problem of bending and distortion. In addition, these procedures are excessively time consuming, and the fibers are not observable during the cutting process. A small microtome has been developed by Hardy³ with which a person, after some experience, can make a cross section in a very short period of time. However, the instrument, being intended primarily for the study of fibers in the textile industry, requires the use of a comparatively large bundle of fibers from which a great many sections are obtained. Of course, such an instrument is unsuited for use in a criminalistics laboratory where the quantity of material available for study is always limited.

Mathiak⁴ described a method of embedding hairs in a celluloid-

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1. Smith, S., and Glaister, J., Recent Advance in Forensic Medicine, Philadelphia, P. Blakiston's Son and Co., 1931, p. 87; Roberts, J.A.F., "A Method of Preparing Sections of Mammalian Hair," Textile Institute Journal, 14, T 114, 1923.
2. Hotte, G.H., "The Fiber Candle Improved to Permit Quicker and Better Preparation of Specimens for Microscope," Textile World, 85, 1063, 1935.
3. Hardy, J.I., "A Practical Laboratory Method of Making 'Thin Cross Sections of Fibers," Circular No. 378, U.S. Dept. Agr., 1935.
4. Mathiak, H.A., "A Rapid Method of Cross Sectioning Mammalian Hairs," Journal of Wildlife Management, 2, 162, 1938.

acetone medium on a strip of balsa wood. After adequate drying, a section was cut freehand through the cellulose acetate layer, hair, and wood base, all of which was then mounted on a slide. The extraneous wood material has been found to be of no particular value as claimed by Mathiak but, in fact, led to the difficulty of separating the hair sections from the balsa fibers. Sectioning of various regions of the same hair was difficult as the wood block was cut in pieces with every initial cut of a series. Freehand cutting was also not desirable because of the thickness and lack of reproducibility of the resulting sections. It was said that sections varied from 0.3-1.0 mm. in thickness.

The wood, on the other hand, provided an excellent, firm support for the hair and was conducive to true right angle cuts. The technique described here combines embedding on a wood base with use of a hand microtome which allows sections of 0.01-0.015 mm. in thickness, or thicker, to be obtained consistently even by the novice. Wood is eliminated from the initially cut material, and sections may be made rapidly from proximal, medial, or distal regions of the same hair which is always visible. Furthermore, the continuity of the whole is not destroyed by cutting of the balsa block.

EXPERIMENTAL

Strips of balsa wood 1 cm. wide and 3 inches long are cut from 1/4 inch thick balsa planks. On one of the centimeter-wide surfaces a single layer of cellulose-acetate glue is applied and spread out evenly to cover completely the wood strip in its central portion. Slow drying "Comet" model airplane cement has been found to be readily available, convenient to use, and quite satisfactory as an embedding medium. While the film of cement is still wet the hair or hairs to be sectioned are laid on it parallel to the long axis of the balsa base. Then, as the film becomes tacky, a second layer of the embedding medium is applied and spread to completely cover the hairs. The block, labeled on the reverse side, can then be placed in a closed container, such as a small box, to dry slowly.

Two precautions should be observed in the embedding procedure. No more than the necessary two layers of cement are to be applied because thicker films are much more difficult to cut. Also, it has been found advisable not to accelerate the drying process since numerous and abnormally large bubbles are thereby formed which if they surround the hair, tend to decrease the tenacity with which it is held. Upon drying, small bubbles are seen throughout the embedding medium,

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but they do not ordinarily interfere with the quality of the final sections. Sections may be cut within $\frac{1}{2}$ hour if necessary, but they are thick and wedge-shaped since the embedding cement is not completely dry, tending to yield in front of the cutting edge. At least twelve hours, or over night, is recommended as a satisfactory drying time to insure the film being *completely* dry and hard.

The hair to be sectioned must be clean and dry before embedding. This is achieved by thorough washing in acetone or alcohol-ether mixture and drying on filter paper in the open air for a short while. The process need not take longer than a few minutes. If a number of hairs of a known source are to be embedded on the same block, they may be oriented, while drying, so all proximal or distal extremities are in the same direction. This procedure facilitates handling of the specimens while being affixed to the block.

After thorough drying, the wood strip carrying the embedded hair is placed in the specimen clamp of a table model hand microtome, e.g. American Optical Model 900. The microtome is then adjusted until the portion of the hair that is desired for sectioning is even with the blade holder guides. A razor blade holder, designed for the instrument, is used always with a fresh, sharp blade. The cut is made, with a short slicing motion, through the embedding layers and hair, *but only* as far as the underlying wood base. The next cut is made in the same fashion after advancing the block the amount desired. As many cuts may be made successively as required.

In the event that another portion of the hair must be sectioned, the block is adjusted to the region of interest and the cutting process is repeated. As the fiber is always in view through the cellulose-acetate layer it is very convenient, for example, to obtain sections close to the ends, in the center, or any other area of the hair. The severed but unsectioned parts of the hair remain attached to the uncut block in their original relative position and may be recovered by dissolving away the lacquer with acetone. The block has suffered little damage and may be resurfaced with sand paper and used repeatedly.

The hair sections, set in a slide of cellulose-acetate 0.5-1.0 m.m. deep and 1 cm. wide, are lifted from the block with a pair of fine-tipped forceps and placed in a small evaporating dish. The lacquer is dissolved with 1-2 ml. of acetone. Upon evaporation of the acetone it will be noticed that the resolidified celloulose acetate has migrated up the sides of the dish leaving the clean, free sections in the bottom. While they are under observation with a stereoscopic binocular microscope, the sections are transferred to a previously albumized slide by touching each with the tip of a sharpened dissecting needle, to which they cling, and then depositing them on the slide. The slide should be inspected briefly under the stereoscopic binocular microscope, and any sections which are observed to be standing on edge can be gently teased into their transverse plane with a needle. An alternate and slightly faster procedure is to separate the lacquer slices and mount them on a slide. However, the lacquer slice surrounding the hair sections interferes somewhat with definition of the contour of the hair.

A permanent mounting medium and cover slip is then applied to the preparation by standard techniques. Clarite "X" dissolved in Xyol has been found to be excellent for the purpose. In addition to its mounting properties the medium infiltrates the hair and clears it nicely for observation.

SUMMARY

The described method of cross sectioning hairs has been found to be well adapted to the requirements of the criminalist. Hitherto, this aspect of hair examination was often avoided because of the lengthy procedures required for embedding and the uncertain results obtained therefrom. This technique, requiring only an hour for sectioning and mounting after embedding, is rapid, simple, and results are consistently reproducible. Materials required are few and readily obtainable, the most expensive item being the microtome. The type utilized, however, is one of the least expensive presently on the market. It is felt that the more extensive use of transverse sections will, in many cases, lead to more positive identifications and comparisons of both human and animal hairs.

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