A NEW METHOD OF MARKING PARAFFIN BLOCKS FOR PURPOSES OF ORIENTATION OR IDENTIFICATION.¹

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THE method described in the following paragraphs is one I have devised and am employing with satisfactory results. It is intended as a simple and rapid means of marking tissues embedded in paraffin for purposes of orientation or identification. Believing that it may prove of some assistance to other microtomists, I append a detailed description.

The procedure consists in embedding in the same paraffin block as the tissue a thin pencil of coloured paraffin wax, having a melting point somewhat higher than that of the embedding mass. Whilst this mass is molten, a length of coloured wax pencil, sufficiently long to project above the block moulds, is placed vertically in the liquid wax in any desired relation to the tissue; this projection of the pencil renders manipulations more easy. The block is then cooled, all protruding paraffin pencil is broken off, and the block is ready for cutting. By this means a clear and definite mark obtains throughout the depth of the block (Fig. 3, *g* and *h*).

The coloured wax pencils are prepared in the following manner: Paraffin wax of a melting point about 10° C. higher than that used in the embedding oven is placed in a porcelain basin and heated over a sand-bath until it melts, the colouring ingredient is then added and the mixture well stirred. Paraffin of this degree of hardness is used because it can be handled as a solid object in the moulds, whilst the embedding paraffin is in a fluid state. The molten mass is then drawn by suction into pieces of glass-tubing about 40 cms. long and 0.5 to 1 mm. in diameter. Before charging these, their inner surfaces must be evenly coated with some thin oil, such as is used for sewing-machines; this is most easily accomplished by running the oil through long pieces of the tubing and allowing them to drain for a few minutes before cutting them into suitable lengths. The object of lubricating the interior of the tube is to prevent the paraffin pencil or rod adhering to the glass and thus producing fracture when attempting its withdrawal.

After charging the tubes they are allowed to cool, then with a file or a diamond a scratch is made about 1 cm. from that end of the tube which was dipped into the melted paraffin; this end is selected on account of the paraffin within the tube being firmly attached to it by the congealed drippings. The tube is then placed upon a table with the scratch or cut turned upwards, and a small piece of thin and moderately soft string is laid beneath the scratch,

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between the table and the tube (Fig. 1), under these conditions a very small amount of pressure, exerted downwards upon the tube a short distance upon either side of the scratch, is sufficient to fracture the glass and to leave the paraffin pencil intact and attached to the short piece of tube. By gently pulling the two pieces of tubing apart the entire casting or pencil of wax may be easily withdrawn. It is then broken off from the attached piece of glass and the grease on it removed by rolling it gently upon a piece of blotting-paper. Pencils so made should be cut into convenient lengths and stored in clean dry test-tubes or other containers.

FIG. 1.—*a*. Scratch ; *b*. glass tubing ; *c*. string ; *d*. table.

FIG. 2.—*e*. Section of plain wax pencil ; *f*. section of cored wax pencil.

FIG. 3.—*g*. Cutting surface of marked block ; *h*. perspective diagram of same block ; *i*. paraffin pencil ; *j*. embedded tissue.

The making of these pencils may be altered slightly, in order to core them with a differently coloured wax. This is done by charging the tubes in the ordinary way and immediately blowing down them ; the down-blast discharges the central mass which has not had time to set, and leaves a relatively thin coating on the inner surface of the tube ; the tube is then recharged from a basin containing a differently coloured wax and the contents allowed to set. The subsequent treatment is the same as that already described (Fig. 2).

For colouring the paraffin, aniline dyes will be found very useful. Many of these are not soluble directly in the melted wax, but if first dissolved in a small quantity of stearic acid, heated just to its melting point, and the paraffin wax be then added to this mixture, the latter will readily take up the stain. The same holds good of certain other dyes besides those of the aniline group. There are several pigments, however, which may be added directly to the heated wax.

In the following list of colours, only those are mentioned which are to be found in most laboratories :—

<i>Black</i> .—Finely pulverised animal charcoal.	{ These pigments may be mixed directly with paraffin wax.
<i>Red</i> .—Congo red ; sudan III.	

<i>Blue.</i> —Victoria blue.	} These dyes must be dissolved in stearic acid and then mixed with the melted wax.
<i>Green.</i> —Victoria (malachite) green.	
<i>Yellow.</i> —Chinolin (quinoline) yellow, also naphthol yellow.	

Generally speaking the colouring should be prominent and intense; light shades are almost invisible in thin sections and consequently are unreliable. By using a series of these pencils in varying combinations of colour, either solid or cored, a very wide range of marking is possible, which could be further increased by casting square or triangular rods in addition to the cylindrical form and combining shape as well as colour.

This method of marking specimens is only applicable for those embedded in paraffin and may be used with benefit in routine as well as in special research work; it is peculiarly adapted as a reconstruction guide in embryological investigations. Should a number of different specimens so marked become mixed through some accident, they may be readily identified.