Chemistry and its Bureau of Investigation are continuously busy in the public interest. It has haled untold rascals before the bar of public opinion; broken up hundreds of shell games.

We unreservedly recommend "Your Money's Worth" as a book that is not only readable and teeming with facts, but as one that will appeal to the physician, both in his professional capacity and also as one of the great army of ultimate consumers of modern merchandise.—*The Journal of the American Medical Association.*

## SCIENTIFIC APPARATUS AND LABORATORY METHODS

### PRE-STAINING IN BOTANICAL MICRO-TECHNIQUE. THE ALCOHOL-XYLOL-SAFRANIN METHOD

A FEW years ago the writer had occasion to imbed some small botanical objects in which it was important to section accurately with respect to the median axis. On clearing in xylol, the material became quite transparent, as so frequently happens, making it almost impossible to see the pieces in the paraffin block, much less to orient the material correctly for cutting. In the ribbon it was again difficult to find the small sections or to ascertain if the material was at all suitable for mounting and staining. The difficulties just enumerated are familiar to all technicians, whether small or large objects are dealt with and, as a result, much valuable time is lost, materials are ruined and the finished product is often thrown into the discard. The writer had read somewhere that it was possible to stain materials in bulk to render them more conspicuous, but details were lacking. Probably it was thought to be altogether too simple a procedure to require further elucidation. Inquiries put to several technicians did not elicit much definite information. This is not surprising, since few workers seem to be aware of the many advantages of pre-staining in micro-technique.

A method of pre-staining was devised which was so generally successful with all kinds of materials and which required so little extra effort that all subsequent imbedding has been done in this way. Not only are imbedding and cutting facilitated, but permanent mounts are also possible without extra labor. The plant parts are killed and fixed in the favorite fluid, and washing, hardening and dehydrating follow in the ordinary way. The clearing is done in alcohol-xylol mixtures, a series of 5, 10, 30, 50, 75 and 100 per cent. xylol in absolute alcohol being generally employed. The stain mixture is inserted in the series in place of the 75 per cent. alcohol-xylol. It is prepared as follows: safranin is dissolved in absolute alcohol to make a saturated solution; 100 parts of the alcoholic safranin are mixed with 300 parts pure xylol. Some of the stain will precipitate out, and the mixture may be filtered, although this is not absolutely necessary. The material is run up through the lower percentages of xylol in alcohol through the 50 per cent. mixture and is then put directly into the safranin mixture, where it is left for 24 hours or longer, depending upon the size and quantity of the material to be stained. It will be seen that the material assumes a deep red color, the fluid at the same time becoming somewhat clear. If much material is to be stained, the original solution may be replaced with fresh stain mixture. There is no danger of over-staining. From the stain mixture the material is run through pure xylol. This will bring down additional safranin as a precipitate, which may be removed by an extra washing with xylol. Embedding proceeds in the regular way. This becomes an easy task, even with small objects, and the pieces are easily seen in the paraffin block. Cutting is facilitated, and in the ribbon the sections stand out clearly. The ribbon may be examined under the microscope and surplus and useless sections may be eliminated with certainty at once. Mounting is done with a minimum of albumen fixative, using no more water than is necessary to smooth out the sections. Any excess water is immediately drained off and the slides are thoroughly dried with gentle heat. Twenty-four hours are not too long a time for drying, and an incubator is best used to eliminate dust and to guard against melting the paraffin. The slides may then be finished. The paraffin is removed with xylol in the ordinary way. At this stage they may be examined under the microscope. The sections will be found to be beautifully stained, and every detail will stand out against a perfectly clear background. A second elimination of unfit material may be made at this time with great certainty, and the only precaution necessary is to keep the slide wet with xylol during the period of examination. The slides may then be finished up by applying balsam and a cover-glass; or they may be run down through the alcohol series, which removes the stain, and any other staining method pursued.

It will thus be seen that this gives a method by which permanent mounts may be made quickly and easily. It is often desirable to make such preparations for temporary class use, and workers in certain fields will find the method adapted to many uses. Mounts thus made have been kept for months without apparent deterioration, the success being apparently determined by the elimination of all sources of water. If a safranin soluble only in alcohol were

possible of attainment, mounts made in this way would probably keep indefinitely. Diaphane has not been tried as a substitute for balsam.

The advantages of the method may be summed up as follows:

(1) Substitution of the stain mixture for 75 per cent. alcohol-xylol is hardly to be considered as an extra effort.

(2) The red objects, no matter how small, are readily visible, even in the hardened paraffin block.

(3) Accuracy of cutting is facilitated.

(4) The cut sections are quite visible in the ribbon and material may be examined superficially for accuracy of cutting, stages desired, etc.

(5) Unfit material is eliminated without further waste of time, and sections of value are not inadvertently thrown away.

(6) Critical examination of material may be made in the stage of removing the paraffin.

(7) Finished mounts may be made at once, without the necessity of going through tedious processes, and especially the individual staining of slides.

(8) Slides so made are fairly permanent.

(9) Slides not intended for quick mounting may be destained and subsequently treated to any other staining technique.

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## A STUDY ON THE LIFE HISTORY OF THE BROAD FISH TAPEWORM IN NORTH AMERICA

RECENTLY the committee on scientific research of the American Medical Association made a grant to the writer in support of a field study on the life history of the broad fish tapeworm, *Diphyllobothrium latum*. This species is a well-known and somewhat serious parasite of man in various regions of the Old World. It was first reported in the United States by Leidy, who studied in 1879 a specimen taken from an immigrant. Other cases which certainly were introduced have been reported from time to time and these records have increased rapidly in frequency within recent years.

The first case in the human host unquestionably infected by larvae bred in this country was reported by Nickerson in 1906 from the clinic of Dr. Parker, of Ely, Minn. The patient was a boy only two years old who had never been out of the state and had never eaten imported fish. In 1911 Nickerson published data on 65 cases from Minnesota, including another record of local infection. Other indigenous cases have been reported by Warthin from Michigan, by Becker from Chicago, by Magath and by Riley from Minnesota, and by Lyon from Indiana. In some districts this species has come to be the most abundant and important human cestode, and this abundance is of very recent origin.

The European form has been introduced into North America many times as more than one hundred cases in man were recorded up to 1922; the list has grown since then though many cases are still unpublished. In fact in certain regions such instances have become too frequent to justify publication. The ova of the parasites were disseminated by sewage systems and thus fishes in connecting rivers and lakes are infected. The history of the parasite at Lake Geneva (Switzerland) is a striking illustration of the way in which the condition is caused and also corrected. No one has as yet shown that the parasite can find here intermediate hosts and the particular small crustacea functioning as such in Europe are rare or unknown here. Moreover, since no accurate examination has been made of the adult tapeworms taken from man here, it may be that the hosts which were infected on this continent really sheltered a new, similar and yet unrecognized species and not the well-known type found in the Old World.

Closely related if not identical species have been reported from other hosts than man in this country, thus by Warthin from the gray fox in northern Michigan, and by Hall and Wigdor from the dog in Detroit. The latter authors regarded the form they described as a new species and named it *D. americanum*. I have myself seen such a tapeworm taken from a dog at Ely, Minn., by Dr. J. E. Thompson. I have also adult tapeworms of this type collected from bears in the northwestern United States and in Alaska. The adult specimens from this continent have not been studied sufficiently precisely to justify a positive statement concerning their specific identity with the Old World species.

The last larval stage, *i.e.*, the form by which the final host is infected, is known as a plerocercoid and occurs in various fish. These plerocercoid larvae are so simple in structure and so imperfectly known that as yet no one can pass upon their relation to definite adult species. I have often found such larval stages of bothriocephalid tapeworms in fish studied in various regions from the Great Lakes to Alaska. Nickerson also records finding such larvae but states distinctly that in the present state of knowledge it is impossible to determine the species to which they belong.

The rapid increase in the number of cases of human infection reported in the United States, the consequent increasing contamination of our streams with probable like increase in infection of fish, and the severe anemia incident to the parasitization of the