

P. S. As this goes to press, November 17, nine of the thyroidless tadpoles are still alive and have increased in size. One has been fed thyroid preparations for 25 days but in very small amounts and at long intervals for fear of fatal results. The hind legs have reached a length of 9.5 mm. and the fore legs are evident beneath the skin. The tail has become greatly shortened and the head is assuming the character of a frog's head. The legs of all the other tadpoles have remained stationary in development at a length of 4 mm. and the tadpoles as a whole show no further signs of metamorphosis. Another tadpole of 29 mm. heavily fed with thyroid, died after 4 days. When compared with a thyroidless tadpole of practically equal body length it was found the intestine of the thyroid fed tadpole had been reduced to a length of 143 mm. as compared with 237 mm., the length of the intestine of the thyroidless tadpole that had not been fed thyroid.

B. M. A.

MICROTECHNICAL METHODS FOR STUDYING CERTAIN PLANT-SUCKING INSECTS IN SITU

A PROBLEM on which the writer has been working for the past year, viz., determining the relation of certain sucking insects to their host plants, has necessitated the development or adaptation of several points of microtechnique which may be of use to other investigators along similar lines. Sectioning insect and plant tissue together has not been attempted often, as the usual methods suitable for one are out of question for the other. It is also necessary to cut quite hard tissues and to employ stains for chitin which also will not dissolve the middle lamellæ separating the plant cell-walls.

The material for study must be fresh. Usually most satisfactory results are obtained if the bottles of killing fluid are taken into the field, the parts of plants bearing the insects cut off with a sharp knife, and immediately immersed. Aphididæ and others of the more active forms must be removed with the part of the plant on which they are feeding and killed before they have time to pull out the proboscis, otherwise their natural positions in

feeding can not be studied. For the gelatine method of embedding, which the writer has used quite extensively, pieces of pine needles, each bearing a coccid at one end, are tied in bundles of ten to twenty, making it possible to get sections of many needles at once; with the use of a killing and fixing agent which penetrates rapidly and easily no difficulty from improper fixing of parts of these bundles is experienced.

Of the killing solutions a variety was tried, Jeffry's¹ proving in most cases the most satisfactory, as the picric acid in it stains chitin. Also it softens hard plant tissues so that it is possible to cut paraffin sections of leaves as hard as *Citrus* without further softening. It may be used hot for twenty minutes or cold for several hours. Care must be taken to wash thoroughly in alcohol or iodine alcohol (30 per cent. alcohol which has been turned to a light wine color by the addition of iodine), otherwise crystals of mercuric bichloride will remain. Carnoy's fluid² also proves successful, particularly with active insects having much secretion of wax, e. g., the aphid *Chermes*, which with less quickly penetrating solutions encloses a drop of air, thus enabling the insect to free its beak before death. Its hardening properties are overcome by thorough washing in absolute alcohol, followed by 95 per cent., 85 per cent., 70 per cent., 50 per cent., 30 per cent. strengths, and then softening in Jeffry's solution.

The gelatine method for embedding³ has been found by the writer very successful for many hard tissues. It deserves extended trial with plant tissues usually considered too hard for sectioning. It is a short method: the material does not require dehydration before its use, therefore hard tissues are not rendered harder than they naturally are. Further, the

¹ Corrosive sublimate, saturated solution in 30 per cent. alcohol, 3 parts. Picric acid, saturated solution in 30 per cent. alcohol, 1 part.

² Absolute alcohol 6 parts, chloroform 3 parts, glacial acetic acid 1 part.

³ Land, W. J. G., "Microtechnical Methods," *Bot. Gaz.*, Vol. 59, May, 1915, p. 400. Chamberlain, C. J., "Methods in Plant Histology," 3d revised edition, p. 128.

natural condition of the structure is maintained, there being but little shrinkage. Cell contents relatively insoluble in water, but soluble in xylol and oils, are not lost. Sections as thin as 10 microns can be cut with ease. However, serial sections can not be cut, nor can some stains be used unless the gelatine is dissolved away after sectioning, which is not easy to do. The method is as follows: Ordinary cooking gelatine is soaked two or three hours, or until it will absorb no more water, then after the excess of water is poured off it is warmed until melted. A temperature of not over 70 degrees Centigrade should be maintained. Part of the liquid gelatine is now thinned with an equal volume of water and the material to be embedded is kept in this dilute gelatine for several hours, during which it must be warm enough to remain liquid. Following this, concentrated gelatine is used similarly for several hours more. The dishes containing the material being embedded should be corked to prevent drying. The material is now cooled in a paper tray coated with paraffin, after which it is hardened for several days in 4 per cent. formalin. The microtome knife must be sharp, with no bevel on the lower side, and set at as great an angle as possible. Either alcohol or water may be used to flood the knife in cutting. Pieces of the gelatine with embedded material are, as a rule, strong enough to be clamped in place in the machine without wooden blocks as supports.

Materials which can not be cut otherwise yield easily to the knife after the use of dilute or concentrated hydrofluoric acid⁴ for one to three weeks, which is followed by thorough washing in water, then the regular paraffin method. Ample time for each stage of the paraffin method to permit dehydration and embedding of the large pieces must be given.

Acknowledgment for many suggestions is made to Dr. L. L. Burlingame, of the botany department of Stanford University.

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⁴ Bailey, I. W., "Microtechnique for Woody Structures," *Bot. Gaz.*, Vol. 49, January, 1910, p. 58.

THE ECOLOGICAL SOCIETY OF AMERICA

A MEETING of the Ecological Society of America was held in the High School building at San Diego, California, on August 10 and 11, 1916, and two joint sessions were held with the Western Society of Naturalists. About twenty-five members were present, the chair being occupied by the secretary-treasurer. Members of the society participated in the biological dinner at the U. S. Grant Hotel on the evening of August 12. On the afternoon of that day the work of the Scripps Institution was demonstrated by members of its staff. On August 13 and 14 the members of the Ecological Society were guests of the San Diego Society of Natural History on a 200-mile automobile trip to the Cuyamaca Mountains and the edge of the Colorado Desert.

Following are abstracts of papers presented at the sessions of the society:

The First Stage in the Recession of the Salton Sea: D. T. MACDOUGAL.

The Trees and Shrubs of the Grand Canyon of the Colorado: ALICE EASTWOOD.

The zones of plant life in the Grand Canyon may be defined by the trees and shrubs which characterize them. The great diversity of environment results in complexities of distribution which offer a promising field for ecological investigation. Fifty lantern slides were shown, made from herbarium specimens of the leading trees and shrubs of the Canyon, collected on the Bright Angel, Hermit and Berry trails.

Results of the Effect of Chaparral and Forest Cover on Meteorological Conditions: EDWARD N. MUNNS.

Records have been taken daily at three stations at the Converse Experiment Station, for three successive years. One station is located in an open cienega, one in a chaparral field, the third in a forest of jeffrey pine, all stations being about 6,000 feet elevation.

The records show the mean annual temperature under the chaparral cover is 2°.8 higher than in the open, and that of the forest 1°.2 higher. More important are the extremes, the mean maximum in the chaparral, being 5°.7 higher and the mean minimum 2°.0 lower than in the open, while the mean maximum under forest conditions is 1°.4 lower and the mean minimum 3°.8 higher than in the open. The mean daily range in the open is 26°.5, that of the chaparral 7°.7 greater, and that in the forest 5°.2 less.

Soil temperatures are greatest in the open, and least in chaparral with a difference of 1°.0 be-