Opinion 99. *Entamoeba* 1895, with *blattae* as type by subsequent (1912) designation, is absolute synonym of *Endamoeba* Leidy, 1879a, p. 300, type *blattae,* and invalidates *Entamoeba* 1895, type by subsequent (1913) designation *hominis* = *coli.*

Opinion 100. Under Suspension of the Rules the genotype of *Spirifer* Sowerby, 1816, is fixed as *Anomia striata* Martin, and the genotype of *Syringothyris* Winchell, 1863, is fixed as *Syringothyris typa* Winchell (= *Spirifer carteri* Hall).

Opinion 101. The technical Latin designations used by Danilewsky, 1891, Annales de l'Institut Pasteur, Vol. 5 (12), pp. 758–782, are not in harmony with the International Rules of Zoological Nomenclature and are therefore not subject to citation or the Law of Priority on basis of said publication.

Opinion 102. A generic name (example *Proteocephalus,* 1858) is not invalidated by the earlier publication of the identical or a similar name of higher rank (example *Proteocephala,* 1828). If *Taenia ambigua* (tod. of *Proteocephalus,* 1858) is congeneric with *ocellata* (tsd. of *Ichthyotaenia,* 1894), *Ichthyotaenia* is a subjective synonym of *Proteocephalus.*

Opinion 103. The type of *Grus* Pallas, 1767, is *Ardea grus* Linn., 1758, by absolute tautonymy. *Grus* is hereby placed in the Official List of Generic Names.

Opinion 104. The following 57 generic names, with type species cited, are hereby placed in the Official List of Generic Names:

PROTOZOA: *Bursaria, Eimeria, Laverania, Plasmodium, Sarcocystis.* CESTODA: *Ligula.* NEMATODA: *Filaria, Heterodera, Rhabditis, Strongylus, Syngamus.* OLIGOCHAETA: *Enchytraeus.* HIRUDINEA: *Haemadipsa, Limnatis.* CRUSTACEA: *Armadillidium, Astacus, Cancer, Diaptomus, Gammarus, Homarus, Nephrops, Oniscus, Pandalus, Penaeus, Porcellio.* XIPHOSURA: *Limulus.* SCORPIONIDEA: *Scorpio.* ARANEAE seu ARANEIDA: *Avicularia, Dendryphantes, Dysdera, Latrodectus, Segestria.* ACARINA: *Cheyletus, Chorioptes, Demodex, Dermanyssus, Glyciphagus, Polydesmus, Psoroptes, Rhizoglyphus, Trombidium.* THYSANURA: *Lepisma.* COLLEMBOLA: *Podura.* ORTHOPTERA: *Blatta, Ectobius, Gryllus, Periplaneta.* ANOPLURA: *Pediculus, Phthirus.* HEMIPTERA: *Anthocoris, Nabis, Notonecta, Reduvius, Triatoma.* DERMAPTERA: *Forficula.* SUCTORIA s. SIPHONAPTERA s. APHANIPTERA: *Pulex.* MAMMALIA: *Cercopithecus.*

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

THE SUBCUTANEOUS LYMPH SAC OF THE FROG AS A CULTURE CHAMBER

MUCH of the tedium involved in maintaining tissue cultures may be avoided by taking advantage of the natural culture chamber afforded by the subcutaneous lymph sacs of the frog. The large ventral lymph sac is especially suitable for this purpose. A slit cut in the ventral skin in the region of the sternum makes it possible to slip excised bits of various frog tissues into the ventral lymph sac, where conditions are favorable for a continuation of living processes in the explanted tissue. The lymph of the living host serves as an aseptic and nutritive medium for the explant, and the explanted tissue may be left undisturbed until the conclusion of the experiment.

Lymph-sac cultures of integument may be maintained for two months or more, but the differentiated cells of liver and kidney undergo early disintegration. Explants of stomach wall and lung wall are more often successful in the lymph sac than those of liver or kidney but do not persist as long as do cells of integument.

In cases where small pieces of integument were inserted into the ventral lymph sac with their epidermis in contact with the subcutaneous surface of the skin of the host, sections taken from a series of such cultures show that the epidermal cells move out from the cut edges of an explant and migrate along the surface of a lymph coagulum which forms between the explant and the subcutaneous surface of the ventral skin of the host. At about forty-eight hours after operation, the migrating cells complete the formation of a vesicle, the wall of which consists, in part, of a newly formed epithelial layer and, in part, of the original explant. The tendency of pieces of integument to form vesicles when cultivated in the frog lymph sac was reported by Winkler (1910), who made no attempt to account for the phenomenon.

About thirty days after operation, sections show that growth of the dermal cells of the explant has begun. This dermal growth continues until the vesicle mentioned above becomes completely invested with dermis. Beginning about three weeks after operation, the subcutaneous blood vessels of the host skin produce branches which invade the newly formed dermis of the vesicle. In later stages up to fifty-five days, these invading blood vessels are filled with normal red blood corpuscles, which fact indicates that these vessels are active in connecting the vesicle with the cutaneous circulation of the host. How long such a parasitic existence will persist can be determined only by further experimentation.

Cultures of frog integument in the lymph sac are equally successful, whether the explanted tissues and the host are of the same or of different species of frog. At least this is true in cases where the two species used are *Rana pipiens* and *Rana clamitans.*

This method of tissue culture fails to afford opportunities for direct observations upon living cells, but

it eliminates many of the difficulties attendant upon experiments involving hanging drop cultures.

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A MICRO-TECHNIQUE FOR OBSERVING OIL PENETRATION IN CITRUS LEAVES AFTER SPRAYING

THE rapidly increasing use of white oil of high viscosity in spraying for the control of scale pests of citrus has been followed in many instances by injurious effects, such as fruit drop, leaf drop, dead-wood, reduction of bloom and reduction of crop, etc.; hence the need for a careful study, by means of the microscope, of the penetration and disposition of oil within the plant tissue.

The problem presented several difficulties. The exigencies of the case precluded the employment of the ordinary solvents used in histological technique. It was necessary to fix or clear the specimen without disturbing or dissolving the oil. Consequently, the use of alcohol, xylol or any other of the essential oils was prohibited, as was also the paraffin method of imbedding which necessitates infiltration with chloroform or ether.

It was desirable to examine both flat sections, cleared and stained, and also fixed cross-sections. This has been accomplished by the use of an aqueous solution of pyridin to dissolve chlorophyll, and Oil Red O[1] dissolved in aqueous-pyridin as a stain.

The technique is substantially as follows:

For Flat Sections:

Immerse specimen in a 60 per cent. aqueous solution of pyridin (60 parts pyridin to 40 parts distilled water).

Heat over water bath. When discolored pour off and refill. Repeat till solution remains clear and specimen becomes transparent. (This can be conveniently done with the use of a small shell vial inserted through the center of a flat cork and floated on water bath.)

Immerse in saturated solution of Oil Red O in 70 per cent. pyridin (70 parts pyridin to 30 parts distilled water) for 24 hours.

Differentiate in 50 per cent. pyridin, until stain ceases to stream. (This takes but a few minutes.)

Wash in running water one hour or more.

Pass through, first glycerine water (equal parts). Second glycerine.

Clear in carbol-glycerine (1 part carbolic acid to 2 parts glycerine). Heat gently in watch glass and observe carefully under dissecting microscope till clear. Specimen should be turned under side up, when oil drops can be distinctly seen.

Pass through glycerine again. (This is important and prevents clouding on the slide.)

Mount in glycerine jelly. Allow to harden and seal with clear Duco.

For Cross-Sections:

Fix in chrome-acetic acid 48 hours.

Wash in running water.

Immerse in 5 per cent. formalin 1 hour.

Wash in running water.

Immerse in 50 per cent. pyridin 10 minutes.

Stain in saturated solution of Oil Red O in 70 per cent. pyridin 24 hours.

Differentiate in 50 per cent. pyridin until color ceases to stream. (Watch carefully.)

Wash in running water.

Section in pith. (The freezing method might be used to advantage but has not been tried.)

Pass through, first glycerine water (equal parts). Second glycerine.

Mount in glycerine jelly and seal with clear Duco.

The oil stains a bright orange to deep red, depending upon the length of time allowed for the staining process. Heavy oils take longer to stain than light. Essential oils, lipoids and other fatty bodies, as well as cutin, also stain but are readily distinguished from the oil. Essential oils are confined to certain well-defined oil cells; they do not occupy the intercellular spaces. Lipoids and other fatty bodies stain deep scarlet, almost black. Cutin stains yellow and, if oil soaked, orange to red.

By employing this technique the writer has been able to observe the penetration of oil into the leaf, its translocation through the vascular system into the stem and across the medullary rays to its final deposition in the storage cells of the pith and old wood fibers. Oils of high viscosity choke the vascular system, to a greater or less degree depending upon the amount of oil, for an indefinite period of time.

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[1] F. Proescher, "Oil Red O a Rapid Fat Stain," *Stain Technology*, Vol. 2: 60.

SPECIAL ARTICLES

ON THE STRUCTURE OF THYMONUCLEIC ACID

THE plant nucleic acid is regarded as a tetranucleotide, each nucleotide being composed of phosphoric acid, a sugar (ribose) and a nitrogenous component. The evidence for this theory of structure is complete, inasmuch as it was possible to decompose the nucleic acid into the individual nucleotides, and each of the nucleotides into phosphoric acid and the complex consisting of the sugar and a base.

For the thymonucleic acid an analogous structure was suggested. The evidence, however, was incomplete, since it was impossible to decompose by chemical means the thymonucleic acid in such a manner