[14] Noble, G. K., *The Biology of the Amphibia*, 1931, 485–86.

[15] *Liopelma* was first described by Fitzinger in 1861 (*Verh. Ges. Wien.*, **11,** 218). Different views have been held about its affinities from time to time. Fitzinger regarded it as closely related to *Telmatobius peruvianus*; Boulenger (1882) placed it in the family *Discoglossidæ*; Nieden (1923) places it in *Cystignathidæ*; Noble, although at first agreeing with Boulenger, later (1924) institutes the family *Liopelmidæ* for its reception. Amongst the recent work on the genus, mention might be made of Wagner's "*Liopelma* studies Nos. 1 and 2" (*Anat. Anz.*, 1934, Bd. 79, Nr. 1/4, 5/6).

[16] Described first by Stejneger in 1899 (*Proc. U. S. Nat. Mus.*, **21,** 899); Regarded by Nieden (1923) as a member of the family *Discoglossidæ*. Its vertebral column is opisthocœlous (Noble, 1922). De Villiers' papers on this genus (*Nature*, 1933, 693; *Anat. Anz.*, 1934, etc.) are interesting contributions on its anatomy.

[17] Lataste, F., "Etude sur le Discoglosse," *Actes Soc. Linn. Bordeaux*, 1879, **33.**

[18] Blanchard, R., "Remarques sur la Classification des Batraciens Anoures," *Bull. Soc. Zool. France*, 1885.

[19] The other families deleted by Noble (1922) are: *Dendrobatidæ*, *Ceratobatrachidæ*, *Genyophrynidæ*, *Hemiphractidæ*, *Amphignathodontidæ*, *Dendrophryniscidæ* and *Dyscophidæ*.

[20] Noble, G. K., *The Biology of the Amphibia*, 1931, 496.

[21] Noble, G. K., "The value of life-history data in the study of the evolution of the Amphibia," *Ann. Acad. Sci., New York*, **30,** 111. The name *Polypedatidæ* is based on the genus *Polypedates*, which had better be called *Rhacophorus* (see foot-note 10, above). Perhap the family should be called *Rhacophoridæ*.

[22] Nieden, Fr., "Anura I and II", *Das Tierreich*, Lief. 46 (1926) and 49 (1926); Ahl, E., "Anura III," *Das Tierreich*, Lief. 55 (1931).

[23] Goodrich, E. S., *Studies on the Structure and Development of Vertebrates*, London, 1930, xxi.

[24] Versluys, J., "Amphibia" in *Handwörterbuch der Naturwissenschaften*, 1931, 296–97.

[25] Werner, Franz, "Dritte Klasse der Craniota. Dritte und zugleich letzte Klasse der Ichthyopsida. Amphibia Lurche." Kukenthal's *Handbuch der Zoologie*, Bd. 6, zweite Halfte; zweite lief., p. 20.

[26] Gadow, H., "Amphibia and Reptiles", *Camb. Nat. Hist.*, 1901, **8,** 19.

[27] He is referring to the species of the Genus *Megalophrys*.

[28] Boulenger, G. A., "A Revision of the Oriental Pelobatid Batrachians (Genus *Megalophrys*)," *Proc. Zool. Soc. London*, 1908, 408.

A Note on Section Cutting of Insects.

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IN Bolles Lee's (1928, p. 510) Microtomist's Vade-Mecum, it is stated that the sectioning of insects is a grim business. Numerous methods of microtomy of insects are reported but none can claim to have a wider range of application (Kennedy, 1932, p. 40), nor is suitable for routine work. It becomes therefore, increasingly, difficult for insect anatomist or histologist in selecting a fixative well suited for a particular material, without the laborious task of giving trial to several of the known methods. Eltringham (1930, p. 93) remarks that there is no satisfactory method of softening chitin without at the same time destroying the structure of the softer internal tissues.

The following fixative which I prepared, gave me satisfactory results in my investigations of the anatomy and histology of insects such as collembola, ants, beetles, etc., perfect sections of the entire insects being obtained as will be seen from photomicrographs given in some of my papers cited in the reference. To enable the beginners interested in the study of insect morphology, to do away with some of the common difficulties which generally hamper the progress of work, I give below the method which we have adopted in our laboratory.

1. Specimens are fixed in the following mixture overnight:

Saturated solution of picric acid in 90% alcohol	..	75 parts
Formalin ..	..	25 „
Strong nitric acid	..	5 „

This fixative as I have already reported (1930, 1932) is a modification of Bouin's fluid, the acetic acid being replaced by nitric acid which it will be noticed had been used before by previous workers as a constituent of some of the fixatives for softening of the chitin in the insect's body. This fixative is useful for embryological as well as for general histological work; and sections are stainable by Hæmatoxylin, Borax carmine and Mallory's mixture (Saüra-fuchsin, Orange G, and Wasserblue). It is, however, inapplicable for cytological investigations; it causes clumping of the chromosomes. The specimens readily sink in the fluid which fixes and softens chitin without damaging the softer tissues. Specimens can be left in the fluid for 12–24 hours; but where a longer time is required for softening chitin that is strongly developed, corrosive sublimate may be added to this mixture so as to prevent maceration of tissues, or the specimens be primarily fixed for a short time in any other fluid such as Bouin's and secondarily transferred to this fluid, as double fixatives are sometimes found advantageous. Where quick penetration is required, suction pump may be used at the time of fixation, or punctures should be made in the specimens if they are very large in size. Often alternate heating below 60°C. and cooling of the fixing fluid containing specimens, are sufficient for driving out air from the tracheæ. Specimens which float in the fluid even after the above treatment should be rejected. In case of heavily chitinised insects it is safer to employ newly emerged specimens.

2. Washing is done in 70% alcohol and dehydration by passing through 90% to absolute alcohol, several changes of alcohol being given.

3. Clearing is done in Cedar Wood Oil.

4. If the specimens are not very strongly chitinised, paraffin sections can be taken, but it is always safe to resort to double embedding although the latter process takes a longer time varying from two to six weeks. In case of double embedding the specimens are soaked for a minute or two in clove oil, and are transferred to a thin and then to a thick solution of celloidin dissolved in clove oil.

5. The celloidin mass containing the specimen, after hardening in chloroform and next in xylol, is transferred to the molten paraffin in the oven. The procedure is the same as in general histology. We prefer paraffin of melting point of 58–60°C. and contrary to orthodox views, a longer bath in the molten paraffin. Hard chitinous substances are cut thicker than 10μ, and to ensure a continuous ribbon the block is coated with a thin layer of soft paraffin. A sharp razor is essential for the success.

6. A difficulty often experienced is that the chitinous parts become loose and float away from the slides as soon as sections are passed from xylol after removal of paraffin, to the absolute alcohol, or, to water after descending grades of alcohol, even when the slides were properly smeared with Mayer's albumen, and good care had been taken in flattening and drying sections. This can be avoided by coating the sections after removal of paraffin by xylol, with a very thin layer of celloidin, by dipping the slide in a thin solution of celloidin dissolved in absolute alcohol and ether. The sections are passed to the descending grade starting from 90% alcohol. In the upgrade passage of dehydration after staining, the sections are quickly passed through absolute alcohol, and clearing is done either in creosote or in a mixture of clove and cedar wood oils in equal proportions. The sections are immersed in xylol before mounting in canada balsam.

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