

32 microns in width and compares well with 0.1 mm. for length recorded for corn embryos aged 4 days (Randolph, 1936). Embryos from 4 days up to 7 days old kernels were similarly excised and all of them cultured in Tukey's general purpose medium (Tukey, 1934) to which were added certain active growth-promoting ingredients indicated in the formula given below:

10 grams KCl	} To 1½ grams of this salt mixture, add 6½ grams of agar, 25 grams of sucrose and 1 litre of water.
2½ " CaSO ₄	
2½ " MgSO ₄	
2½ " Ca ₃ (PO ₄) ₂	
2½ " FePO ₄	
2 " KNO ₃	

*Proportion of Physiologically-Active
Ingredients*

0.2 mg. of adenine per litre of prepared solution.

20.0 mg. of ascorbic acid per litre of prepared solution.

25.0 mg. of succinic acid per litre of prepared solution.

3.0 mg. of glycine per litre of prepared solution.

0.1 mg. of nicotinic acid per litre of prepared solution.

0.5 mg. of pantothenic acid per litre of prepared solution.

0.2 mg. of vitamin B6 per litre of prepared solution.

The Tukey's medium was slightly modified by substituting 1 per cent. glucose with 5 per cent. sucrose, as sucrose is found to promote better embryonic growth in corn than glucose (Haagen-Smit, 1945). The culture bottles were kept in an incubator whose inside temperature was kept at 31° ± 1° C. The 3 to 6 day-old embryos were like tiny specks, making it hardly

TABLE I

*Embryos placed in the culture medium on
Sept. 11, 1947*

Date of measurement	Length of shoot mm.	Length of Root mm.
13-9-1947	2.0	1.9
14-9-1947	2.0	2.0
15-9-1947	2.1	2.0
16-9-1947	2.2	2.3
17-9-1947	2.3	2.4
18-9-1947	2.3	2.4
19-9-1947	2.3	2.4
20-9-1947	2.3	2.4
21-9-1947	2.3	2.4
22-9-1947	2.3	2.4
23-9-1947	2.3	2.4

possible to measure their sizes with reasonable accuracy. The 7 day-old embryo, however, was conspicuous and large enough to measure. The growth measurements for this are recorded in the table below.

The figures are averages for 5 embryos. It will be seen that the growth is very slow and retarded. At the end of the 5th day of culturing the growth, however, stopped. It was evident that the embryos of very young age, i.e., anything less than ten days, required some growth-promoting substance other than those supplied to the medium in the present experiment.

Discussion.—In this experiment it is evident that the young embryos aged 3 to 7 days were not able to mature because they lacked certain specific embryo factors necessary for further growth at this stage. It is also clear that the 7 day-old embryo was able to grow a little because it was fast developing to be autotrophic, but still wanting in certain growth hormones needed for full development. The physiologically active substances contained in the culture medium were perhaps able to supplement this deficiency to a certain extent but not fully. A similar case has been reported by White (1932). He was able to grow an embryo of *Portulaca oleracea* measuring only 0.12 mm. to a size of 1.84 mm. by adding a fibrin digest to his culture medium. The embryo, however, was unable to grow further at the end of the third week.

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1. Haagen-Smit, A. J., and others, "A Method for culturing of excised immature corn embryos *in vitro*," *Sci.*, 1945, **101**, 234. 2. Randolph, L. F., "Developmental morphology of the Caryopsis in maize," *Journ. Agr. Res.*, 1936, **53**, No. 12, 881-916. 3. Tukey, H. B., "Artificial Culture methods for isolated embryos of deciduous fruits," *Proc. Amer. Soc. Hort. Sci.*, 1934, **32**, 313-22.

THE LIFE-CYCLE OF *MONILIFORMIS* *MONILIFORMIS* (BREMSER, 1811), ACANTHOCEPHALA

ALTHOUGH morphological and taxonomic studies have been made on various Acanthocephala during the last three decades, little is known about their life-cycle. The development of *Macracanthorhynchus hirsutinus* of pigs in beetle larvæ was described by Meyer (1931), and its life-history

was traced in the intermediate and definitive hosts by Kates (1943, 1944). In 1941, Burlingame and Chandler, and in 1946, Moore showed that the cockroach *Periplaneta americana* plays the role of the intermediate host for *Moniliformis dubius*, parasitic in rodents. *Moniliformis moniliformis*, which is also a normal parasite of rodents and occasionally the dog and man, is conveyed through the intermediation of the beetle, *Blaps mucronata* (Grassi and Calundruccio, 1888) and also the cockroach, *Periplaneta americana* (Seurat, 1912, and Southwell, 1922). Its occurrence in India has been recorded by Van Cleave (1925) and Bhalerao (1935). An investigation of the complete life-cycle of this thorn-headed worm was therefore considered useful.

Infective larvæ (Acanthellæ) were obtained from the body-cavity of naturally infected *P. americana* and fed in fresh condition to worm-free laboratory-bred rats. The rats were maintained under controlled conditions and autopsied after varying intervals of 1, 6, 12, 18, 41, 59, 62, 72, 128 and 147 days, and the parasitic stages were recovered and studied. In order to precisely determine the period when maturity was attained, the fæces of the infected rats were examined every day for the eggs of the worm, and it was found that in some cases the eggs appeared after 22 days and in others after periods extending up to 38 days. Thus it was observed that the pre-patent period ranged from 22 to 38 days. In one instance, the eggs continued to appear in the fæces for 130 days. This patent period indicates the duration of the fecundity of the worms. The size and structure of the worms at different ages, the ratio between the number of larvæ ingested and of adults recovered, the sex-ratio, and their location in the host-intestine, have been studied in detail.

When full-grown, the males attain a length of 32-106 mm., whereas the females measure 69-230 mm., and as many as 6500 eggs were expelled with the fæces of a rat in a day.

The fertilised egg (Fig. 1), when extruded, has four envelopes and an embryo with numerous spines. Further development is possible only when the insect swallows the fæces containing these eggs. Inside its gut, the envelopes burst and the acanthors emerge. These make their way out of the gut and develop into the acanthella (Fig. 2) in the hæmocœle. This development inside the cockroach was experimentally studied

by feeding laboratory-reared specimens of *P. americana* with the eggs of the worm. The exact duration of acanthor and acanthella stages was timed through 5, 7, 14, 21,

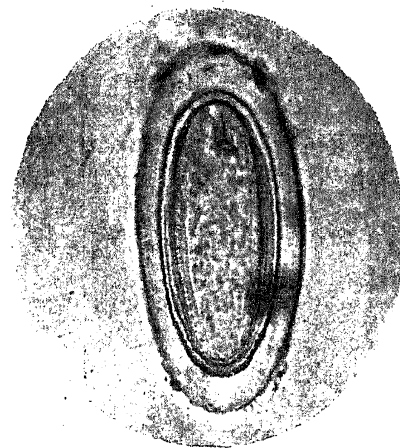


FIG. 1

Photomicrograph of egg of *M. moniliformis*, fresh from fæces of experimental rat. $\times 330$.

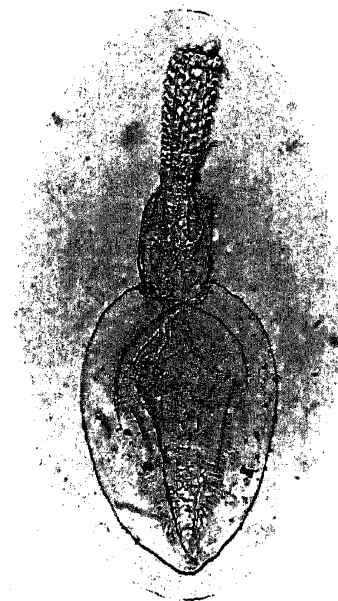


FIG. 2

Photomicrograph of infective acanthella of *M. moniliformis* from body-cavity of *P. americana*, without enveloping cyst. $\times 30$.

28, 35, 42 and 50 days after ingestion of eggs; of these, the first two yielded acanthor stages from the gut, and the rest pre-acanthella and acanthella stages from the body-cavity. The development of the embryo into the acanthor, its migration into the body-cavity, and the morphological changes involved in its development into an acanthella have been followed. The infective acanthella is enclosed in a delicate cyst and has a well-defined organisation in

which even sex-differentiation has taken place.

A detailed account of the above, a discussion of epidemiological considerations such as the viability of eggs and acanthellæ *in vitro*, the mode and conditions of transmission, the intensity of infections as assessed from the insect-host over a period of many months, and the possibility of other arthropod and vertebrate hosts acquiring the infection will be described in a fuller paper.

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April 28, 1949.

Bhalerao, G. D., *Imp. Council of Agric. Res. Sci., Monograph No. 6*, 1935. 2. Burlingame, P. L., and Chandler, A. C., *Amer. Journ. Hyg.*, 1941, **33**, Secn. D., 1. 3. Grassi, B., and Calundr-uccio, S., *Journ. Roy. Micr. Soc. Lond.*, 1888, **8**, 739. 4. Kates, K. C., *Amer. Journ. Vet. Res.*, 1943, **4**, 173; 1944, **5**, 166. 5. Meyer, A., *Zool. Anz.*, 1931, **93**, 163. 6. Moore, D. V., *Journ. Paras.*, 1946, **32(3)**, 257. 7. Seurat, L. G., *Compt. Rend. Soc. de Biol.*, 1912, **72**, 62. 8. Southwell, T., *Journ. Paras.*, 1922, **9**, 99. 9. Van Cleave, H. J., *Proc. Acad. Nat. Sc. Phila.*, 1925, **76**, 279.

A MODIFIED METHOD FOR THE ESTERIFICATION OF SOME POLYHYDROXY AROMATIC ACIDS

It is known that esterification of acids can be carried out with alkyl sulphates or alkyl iodides using alkali hydroxide or carbonate in presence of suitable anhydrous solvents. In case of hydroxybenzoic acids, however, it is likely that this method may simultaneously lead to partial etherification also. The Fischer-Speier method using alcohol and concentrated sulphuric or hydrochloric acid also fails in case of some polyhydroxybenzoic acids. Thus, *o*-orsellinic acid has been esterified only by the diazomethane method,¹ and the ethyl ester had not yet been prepared from the acid. *p*-Orsellinic acid has been esterified with diazomethane and also by the action of methyl and ethyl iodides on its silver salt.² The usual catalytic method fails in these cases probably because of the ease of decarboxylation of these acids, which may be taking place due to the temperature of the reaction or the presence of the acid.

A new method has now been devised

where the esterification is carried out in a dry medium, using a neutral substance like sodium bicarbonate. By this method the methyl esters of both the orsellinic acids were prepared in high yields by refluxing for ten hours in dry acetone with sodium bicarbonate (1.25 mols.) and dimethyl sulphate (1.25 mols.). Excellent yield of the ethyl esters were obtained by similar method using diethyl sulphate (1.25 mols.) or ethyl iodide (3 mols.). It was also observed that even if excess of alkyl iodide was used, the hydroxy groups were not attacked.

α - and β -resorcylic acids also gave good yields by this method; benzoic acid itself however, gave poor yields.

This new method of esterification is a general one and would be particularly useful for some acids for which the catalytic method cannot be used. It is a good substitute to the diazomethane method having an advantage over it, that it is more simple and that esters other than the methyl can also be prepared. Moreover, it has been found to give good results even with small amount of acids.

A detailed account of the work will be published elsewhere.

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Bombay,
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1. Herzig, Wenzel and Kurzweil, *Monatsh.*, 1903, **24**, 895. 2. Robertson and Robinson, *J. Chem. Soc.*, 1929, 2199.

SUGARCANE \times BAMBOO HYBRIDS

RESEARCH work by Doctors Avdulov and Prat as also by Dr. C. A. Taylor of the Cornell University, Ithaca, New York, would appear to indicate a close relationship between the Bamboo and the Panicoid grasses to which the sugarcane belongs. When the sugarcane bamboo hybrids were first effected by me at Coimbatore in 1936 there were certain Botanists who doubted the possibility. Subsequent work on the Chromosome numbers and the morphological and the histological characters of the F₁ Hybrids at Coimbatore appear to confirm the nature of the hybrids. In the work mentioned above there is further confirmation of the possibility of hybridisation between the above two widely different genera of plants.

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