size of the erythrocyte is 13 μ × 8 μ. Few erythroblast cells were also noted in *C. catla*. Their percentage varies from 0·8 to 2·2 with an average of 1·6%. The average size is 9 μ × 7 μ. The presence of erythroblasts has been reported only in *Acrossocheilus hexagonolepis*[8].

The total number of W.B.C. in *C. catla* varied from 7,400 to 11,500/cmm of blood with an average of 9,241. The male fishes have a higher count (9,816) than the females (8,660).

The following five types of leucocytes with corresponding percentages given in the parenthesis were noted in *C. catla*: Thrombocytes (30), Neutrophil (18), Lymphocytes (32) and Eosinophil (16). The nucleus of the thrombocytes is large and is surrounded by a thin rim of cytoplasm. The cytoplasm of eosinophil is pinkish in colour and is filled with numerous small and rounded granules.

The percentage of basophil in *C. catla* is 4 which is much less than that of *A. hexagonolepis*[8] but others[1-7] failed to notice any basophil cells in their studies of the blood of the fresh-water Indian air-breathing fishes.

Thanks are due to Dr. H. Choudhri, Officer-in charge, Central Inland Research Sub-Station, Cuttack, for kindly supplying the fishes and to Prof. B. K. Behura, Head of the Department of Zoology, for providing the laboratory facilities and encouragement.

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ANTIFERTILITY EFFECT OF BIOTIN AND ITS AMELIORATION BY ESTROGEN IN THE FEMALE RAT

The minimum requirement of biotin for maintenance of pregnancy in the rat during its last few days appears to be 3 μg per day[1]. It is not uncommon that hypervitaminosis of various kinds can occur due to excess intake of some vitamins. A high dose of biotin in the rat causes irregularities of the estrus cycle and abnormal infiltration of leucocytes in the vagina in association with atrophic changes in the ovary[2]. However, there are no reports on the effects of an excess intake of biotin on the fertility of the mammal. It was of interest, therefore, to study the effects of acute doses of biotin on the mating behaviour and maintenance of pregnancy in the rat.

Colony bred virgin female rats of the Holtzman strain approximately 3 to 4 months old were selected after observing their normal estrus cycles. A batch of 36 rats thus selected was treated with a dose of 5 mg of biotin per 100 g of body weight dissolved in 0·5 ml of 0·1 N NaOH in two subcutaneous injections (morning and evening). These rats were then kept for mating with males of the same strain in 3 groups of 12 rats each in the order of 7, 14, and 21 days after biotin injections. As and when day 1 of pregnancy was identified by the observance of sperms in their vaginal smears, the females were separated from the males. In each of these 3 groups of pregnant rats, six out of the 12 were sacrificed on the morning of day 15 of pregnancy and the remaining six on day 22 of pregnancy. An untreated control group of 12 rats was run simultaneously and was sacrificed in the same manner. Since it was observed that the equivalent amount of 0·1 N NaOH (0·5 ml) used as vehicles for biotin, did not interfere with the estrus cycle[2], it was felt unnecessary to run a NaOH-treated control along with this experiment. However, towards the end of this experiment after observing some of the results, it was felt relevant to include one more group of 6 rats identically treated with biotin and mated, but given 1 μg of 17 β-estradiol dissolved in 0·1 ml of olive oil daily up to 21 days of pregnancy starting from day six. A higher dose (10 mg/100 g body weight dissolved in 1 ml of 0·1 N NaOH in 4 injections for two consecutive days) of biotin was also tried. All these rats were maintained under standardized light and dark, temperature, humidity and feeding conditions as designed for the best reproductive performance. At autopsy, the foetuses and placentae were dissected out and weighed. The number of implantation sites in the uteri was also counted.

The results of the present study showed that a large number of biotin (5 mg/100 g body weight)-treated rats although mated within 10 days, failed to maintain pregnancy (Table I). In fact most of these biotin-treated rats resorbed their foetuses and nothing but mere implantation sites were present at the end of 21 days (Fig. 1). The foetal and placental weights of the few rats that were able to maintain pregnancy despite biotin treatment were below normal

TABLE I

Effects of biotin on mating and maintenance of pregnancy in the rat

Treatment	No. rats mated	No. rats with foetuses	No. foetuses/rat[a]	No. implantation sites/rat[b]	Foetal weight[a] (g)	Placental[a] weight (g)
5 mg biotin/100 g body weight			Day 14 of pregnancy			
Mating allowed: 7 days after biotin ..	6	6	8·0 ±2·8	11·0 ± 1·0	0·126 ±0·062	0·109 ±0·024
14 days after biotin ..	6	4	5·6 ±1·4	6·5 ± 3·2	0·101 ±0·027	0·191 ±0·074
21 days after biotin ..	6	3	8·0 ±0·0	8·6 ± 1·4	0·163 ±0·002	0·205 ±0·001
Untreated controls ..	6	6	10·2 ±2·0	10·3 ± 1·7	0·721 ±0·132	0·251 ±0·026
5 mg biotin/100 g body weight			Day 21 of pregnancy			
Mating allowed: 7 days after biotin ..	6	2	11·0 ± 0·0	11·0 ± 0·0	3·41	0·501
14 days after biotin ..	6	3	11·0 ± 1·1	11·6 ± 1·2	4·26 ±0·21	0·536 ±0·034
21 days after biotin ..	6	1	9·0	9·0 ± 1·0	5·56	0·614
7 days after biotin + E_2*	6	6	9·8 ± 1·8	10·8 ± 1·8	5·33 ±0·83	0·621 ±0·051
Untreated controls ..	6	6	9·2 ± 3·3	10·12 ± 1·3	5·86 ±0·54	0·602 ±0·153
10 mg biotin/100 g body weight						
Mating allowed 7 days after biotin	None of the 6 rats mated as observed for 2 months					

* = Estradiol – 17 β (1 μg/rat/day starting from day 6 of pregnancy).

[a] = average of the number of rats with foetuses; [b] = average of the number of rats mated.

values (Table I). The biotin-treated rats showed normal body weight gain and apparently maintained good health. When estrogen treatment was given to these biotin-treated rats, all the rats maintained pregnancy (Table I and Fig. 1). The foetal and placental weights also became comparable with those of controls after estrogen therapy. These observations, therefore, directly indicate that the biotin-induced infertility in these animals is due to the deficiency of estrogen. An acute treatment with biotin not only causes disruption of the estrus cycle, it induces atrophy of the corpora lutea and stroma of the ovary[2]. Recently we[3-5] have suggested that continued administration of estrogen to the non-pregnant rat may maintain ovarian progesterone secretion for 21 days, and the interaction of these steroids seems to play a major role in the maintenance of pregnancy physiology of the rat. It seems possible, therefore, that the adverse nature of the pregnancy maintenance following acute biotin treatment is primarily due to inhibition of ovarian estrogen secretion and in its wake inadequate maintenance of the circulatory levels of progestins.

Another interesting point to note in these observations is that the biotin-treated rats did not recover from reproductive inability with the lapse of time, and the higher dose (10 mg) prevented them from even mating as observed for more than two months. It is, therefore, very tempting to suggest that an

acute dose of biotin may eventually lead to a permanent sterility in the rat.

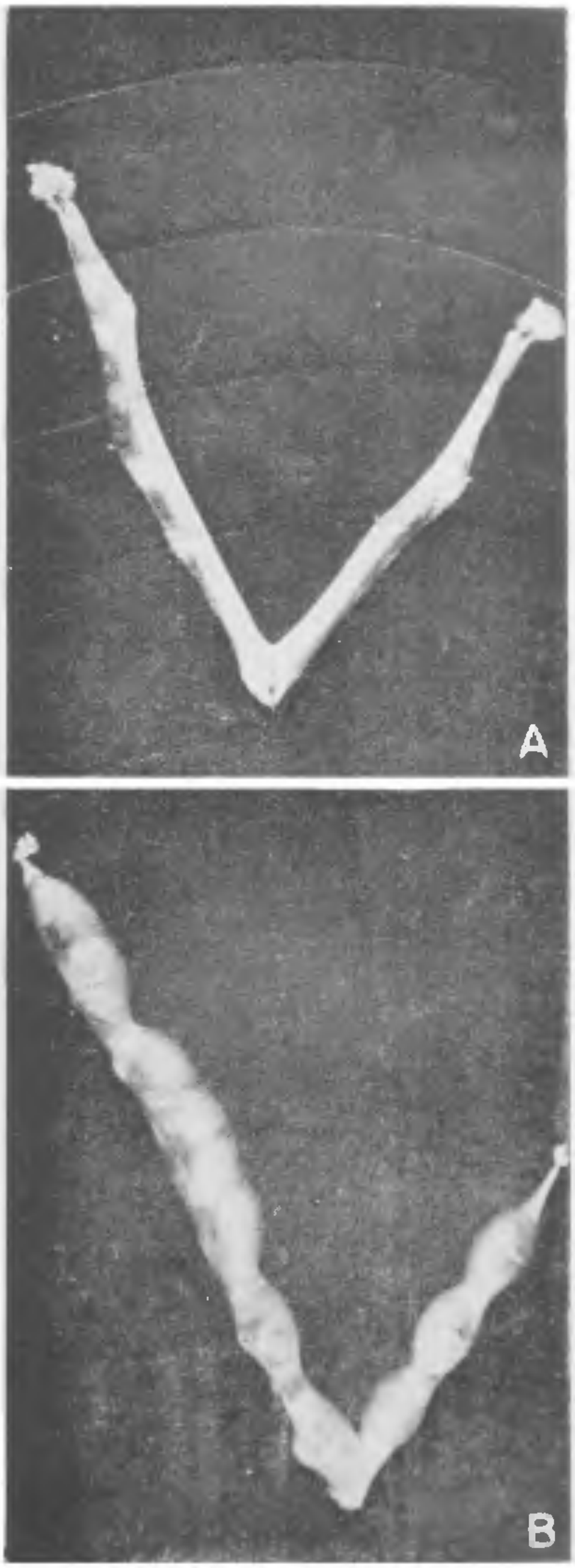

FIG. 1. Effect of biotin alone and biotin followed by estrogen treatment on the maintenance of pregnancy in the rat uterus. A = biotin (5 mg/100 g body weight in two injections). B = biotin (5 mg/100 g body weight in two injections *plus* 1 μg of 17 β-estradiol up to day 21 of pregnancy starting from day six). Photographs were taken on day 22 of pregnancy in the morning.

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A NEW SPECIES OF *XENOSPORIUM* FROM INDIA

THE genus *Xenosporium* was described by Penzig and Saccardo[6] with *X. mirabile* as type, which was later redescribed by Ellis[4]. It is a dematiaceous Hyphomycete producing conidia acrogenously from simple or branched, proliferating conidiophores arising from the repent hyphae. The conidia are characteristically incurved, flattened from side to side, phaeo-dictyosporous, producing "conidioles" (Clements and Shear[1]) or "secondary conidia" (sensu Pirozynski[2] and Ellis[4,5]) from their incurved regions. The "secondary conidia" are usually 2 to many celled while they are continuous in *X. berkelevi* (Curtis) Pirozynski and *X. larvale* (Morgan) Pirozynski. (Despite the common occurrence of secondary conidia in all species of *Xenosporium*, their function in its life-cycle is not very clear). Pirozynski[2] reported six species of *Xenosporium* who also had provided a key for them. In this communication a new species of this genus is being proposed.

Xenosporium shoranoorense sp. nov.

Colonies discrete, effuse, deep brown. Mycelium superficial hyaline to subhyaline, branched, smooth walled, septate, with septa 4·5–7·2 μ apart, 3·4–4·5 μ in diam. Conidiophores arise from creeping hyphae singly or in groups, erect or flexuous, simple, subhyaline to light yellowish brown, proliferating, 2–4-septate, 36·0–62·0 μ long, 3·6–7·2 μ broad. Primary conidia acrogenous, single, oval to cylindric, phaeo-dictyo-aleuriosporous, pale to dark blackish brown, muriform 34·0–54·0 μ long, 20·0–35·0 μ broad, covered by a mantle of cells. Mantle 1-celled in thickness, subhyaline to light brown. Each conidium sometimes on detachment carries with it a portion of conidiophore as a frill which, however, collapses later. Secondary conidia light to blackish brown, globose to subglobose, 0–6-septate, muriform, smooth, 7·2–10·8 μ in diam., produced terminally, laterally and from all over the surface of the mantle.