PRELIMINARY EXPERIMENTS ON THE MODIFICATION OF THE REPRODUCTIVE CYCLE OF AN INDIAN CARP CIRRHINA REBA (HAM.) BY CONTROL OF LIGHT AND TEMPERATURE

by

P.U. Verghese Central Inland Fisheries Research Institute Barrackpore, India

ABSTRACT

Results of an experiment conducted to study the effect of light and temperature on the sexual cycle of *Cirrhing reba* (Ham.), a closely allied species to the commercially important Indian major carps, during 1 August 1966 to 30 June 1967 are presented in the paper. The fish were reared in cement cisterns on artificial food, under different photoperiodic regimes of 8, 14 and 18 hours of constant light per day and varying photoperiods ranging from 4 to 20 hours per day. The Gonadosomatic index was used as a measure of the effect of light and temperature on the reproductive cycle. Fish under the influence of different photoperiods attained functional maturity at different months of the year starting from February. The data indicated that artificial day lengths longer than natural day accelerate the later phase of maturation in the species. Functionally mature fish were subjected to pituitary extract injections and successful breeding is reported.

INTRODUCTION

Investigations on seasonal variation in the reproductive activity of fish and the factors responsible for such variations have revealed that the seasonal changes in the day-length and water temperature are the major factors controlling the reproductuve cycle (Hoar, 1957 and Alm, 1959). Reports on successful attempts to bring about changes in the reproductive cycle artificially by controlling light and/or temperature in different species of fish are available (Hoover, 1937; Hoover and Hubbard, 1937; Hazard and Eddy, 1951; Allison, 1951; Corson, 1955; Comb, Burrow and Bigej, 1959; Harrington, 1950, 1956, 1957, 1959a; Shiraishi and Takeda, 1961; Henderson, 1963; Yoshioka, 1962, 1963 and Shiraishi and Fukuda, 1966). Atz (1957) and Harrington (1959b) have reviewed the literature on the role of environmental factors on the reproduction of fish. These studies have shown that requirements of light and temperature for the activation of the reproductive cycle vary from species to species and from place to place, as the day-length and temperature ranges differ, depending upon the latitude of the locality at which the fish grow. In case of Indian carps, experimental evidences on the influence of light and temperature on their sexual cycle are scanty. Verghese (1967) reported that the spawning season of Cirrhina reba (Ham.) can be prolonged under enhanced artificial light condition.

Proc. Indo-Pacific Fish. Coun., 13(II): 171-184.

As is well known, the induced breeding technique with pituitary extract injections, which has come to stay in India as a source of pure fish seed for culture purposes, has the disadvantage of being confined to the narrow stretch of the natural spawning season (June to end of August) (Alikunhi, *et. al.*, 1964; Bhimachar and Tripathi, 1967). The non-availability of sufficient number of breeders with the required stage of ripeness has also been reported as an impediment to the success of spawn production by induced breeding (Alikunhi, *et. al.* 1960; Chaudhuri, 1960). It will thus be of great practical value if mature carps could be available during any convenient period of the year.

The present study was undertaken to find out the effect of light and temperature on the reproductive cycle of *Cirrhina reba*, a species closely similar to the commercially important Indian major carps in spawning season and breeding habits. The investigation also aims at the possibility of making ripe fish available for induced breeding, through a longer period of the year than the natural spawning season. The following account deals with the preliminary set of experiments covering one complete reproductive cycle of the fish conducted during 1 August 1966 to 30 June 1967.

MATERIAL AND METHODS

Cirrhina reba collected during June 1966 from a local stocking pond were reared in a row of cement cisterns each 180 x 100 x 70 cm, containing fresh water pumped from a tube-well. The water was kept well aerated by bubbling air from a non-lubricated air compressor. A mixture of wheat flour, fish meal, wheat bran and baker's yeast in the ratio 40:20:5 with traces of Terramycin (Oxytetracycline hydrochloride) and cobalt nitrate (50 ppm and 300 ppm respectively) was found to be an adequate feed for the fish.

Artificial illumination was given, using a pair of 40 watt fluorescent day-light tubes mounted at 1.5 m above the water level in the centre of each cistern. Each cistern with the lights was covered with light-proof black cloth. The intensity of light falling on the surface of water in each cistern, was measured with an objective light meter, Type Metrux 2 and was between 200 and 250 lux. Higher water temperature was maintained by automatic immersion heaters.

A group of fish in one of the cisterns without any artificial control of light and temperature was kept as control. Representative samples consisting of a minimum of one male and one female were collected from each group at regular intervals. Weight and length were recorded up to the nearest hundredth of a gram and to the nearest millimeter respectively. The gonads were dissected out and fixed in Bouin's pirco-formol and weighed to the nearest milligram in a sensitive chemical balance, after draining the fixative within the folds of filter paper. The stages of gonadal maturity were determined from the gross morphology and the gonadosomatic index (Gonad weight x 100 divided by gutted weight of the fish) as given by Pickford and Atz (1957), Clemens and Reed (1967) and Bogenschutz and Clemens (1967). During the advanced stages of maturity all the fish were examined individually for functional maturity by hand strippling. Some of the mature fish were induced to breed in the cisterns by administration of pituitary gland extract following the method adopted by Chaudhuri (1963).

RESULTS

The gonadosomatic index (GSI) for males and females was found to give necessary information regarding the progress of gonadal development in the experimental fish. The nature of light periods and temperature ranges employed for the experiment are presented in Tables I and II. The number of fish initially used in each experiment and their range in total length (in mm) are given in Table III. Prior to the introduction of fish to the experimental tanks on 1 August 1966, samples were collected at random and examined for gonadal condition. They were found to be immature virgin forms. The monthly samples taken from the experimental groups till December 1966, did not show well defined variations in the gross morphology of reproductive tissues. Marked differences in size and shape of the gonads and consequent variations in the GSI were recorded from the month of December. Figures 1 and 2 show the monthly changes of GSI of the seven different experimental groups of fish from December 1966 to June 1967. The GSI of immature females during December varied from 0.8 to 1.5 and that of functionally mature ones from 24.3 to 36.9. In the case of males the corresponding values were 0.01 to 0.05 and 0.91 to 1.1. The males in general matured well in advance of the females in all but one experimental group. Numbers of functionally mature males and females and the recording dates are illustrated in Fig. 3. The females were recorded only when they could be recognized externally with bulged, soft abdomen. The GSI of samples killed at this stage were found to be above 20.0 and the ovary filled the entire abdominal cavity. Males were recorded as mature when milt could be expressed by a slight pressure on the abdomen.

Experiment No. 1

This formed the control which received natural day light throughout the experimental period. The temperature of the water varied from 19.0 to 30.0° C. Figs. 4a and 4b show the natural day length curve calculated from sunrise and sunset time for the latitude at Barrackpore, which is between 22 and 23°N, the experimental light regimes and the average monthly water temperature for the experimental period. The gonad development in the control set has been taken as normal for comparison. The fish attained maturity during the normal breeding season of the species in this region, when mature specimens were available in the local fish ponds. All the males available in the group matured during 10th to 30th of May, while females completed the process of maturation between 20th May and 10th June.

Experiment No. 2 - Retarded photoperiod

The fish received 8 hours of light per day from 1 August 1966 to 30 June 1967. The temperature of water varied from 19.0 to 30.2°C. In this experimental group mature fish appeared later than in the control. The first oozing male was noted on 20 May 1967 and mature females were recorded from 10 June 1967. However, more than 50% of the females were recognized as mature only by the end of June.

Experiment No. 3 - Long photoperiod

Fourteen hours of light per day was given to this group throughout the period. The first oozing male was recorded on 20 March 1967 and oozing males

continued to appear till the end of May. The corresponding period for females was 20 April 1967 to 20 May 1967. This was slightly in advance of the earlier two groups.

Experiment No. 4 - Long photoperiod

The duration of light was 18 hours per day. The males matured between 28 February 1967 and 20 April 1967 and females between 10 March 1967 and 10 May 1967. This can be taken as an instance of early maturity as compared to the control. All the fish attained functional maturity before the middle of May, i.e. before the appearance of oozing males in the control set and when no mature specimens of the species were available in Jocal ponds.

Experiment No. 5 - Varying photoperiod

Fish in this experimental group were exposed to a varying photoperiodic regime, which started with 20 hours of light per day from 1 August 1966 and the light period was decreased at a rate of one hour per week till it reached 8 hours per day by 31 October 1966. This 8 hours per day schedule was continued up to 31 January 1967. Again from 1 February 1967 the light period was increased at a rate of half an hour per week. This accelerated light regime was continued till the end of experiment. The length of day gradually falls below 12 hours per day by October and continues to decrease up to the middle of December (Fig. 4a). After remaining constant for two weeks it again rises, reaching the 12 hours per day level by middle of March. The photoperiod employed in the experiment viz., the gradual decrease from August to the middle of October, remaining constant (8 hours per day) throughout the winter months and then increasing from February coincides with the general pattern of the fall and rise of the natural day-length. The gonadal development was affected in quite a different way. Though a few oozing males started appearing from the end of March, the majority of the fish did not mature till the end of June. Female maturity was first recorded by the end of May and the last mature females were detected by the end of June. This particular set was kept in the then prevailing light conditions for a period of two more weeks, during which time 2 more fresh males were recorded as oozing.

Experiment No. 6 - Varying photoperiod

The varying light period employed in this experiment was different from that of experiment No. 5. To begin with the photoperiod started with a reduced quota of light of 4 hours per day. The duration of light was brought up at a rate of one hour per week till it reached 14 hours per day by 31 October 1966. The 14 hours level was maintained till 31 January 1967. Later, as in the previous experiment, the light period was increased gradually and brought up to 20 hours per day by the end of April. Except for the early period, fish under this experiment received more hours of light than natural day light. The effect was quite conspicuous. The first mature females were recorded as early as last week of February. Contrary to the normal experience, oozing males appeared in this group later than the first few mature females. Fish from this group were employed for induced breeding experiments with pituitary extract injections on 16 May 1967, well in advance of the normal spawning season. The fish bred in the cistern water and the larvae produced were reared to maturity in the succeeding season.

Experiment No. 7 - Varying photoperiod with high water temperature

The light period employed for this experiment was the same as that of experiment No. 6. In addition, artificial control of water temperature from 1 September 1966 to 30 April 1967 was also maintained. As in the case of experiment No. 6 the mature females were recorded by the end of February, but oozing males had already been recorded two weeks earlier. Males from this group were the first to mature in the entire lot. All the available males were oozing by the last week of February. The higher water temperature employed during September to April did not give any convincing modification regarding the first attainment of female maturity. However, all the females matured in the month of March, whereas, in the previous experiment a few females took more time to become fully mature. In the males higher temperature seems to have an accelerating effect on maturation, since oozing males appeared in this group nearly 15 days earlier than in the experiment No. 6.

DISCUSSION

Results presented in the above account indicate positively that Cirrhina reba exhibits photoperiodism in its reproductive cycle. The reason for the attainment of early maturity in experiments Nos. 3, 4, 6 and 7 and delay in the maturation process observed in groups 2 and 5 are attributable to the daily duration of light received by each group. As will be seen from the summary of results presented in Table IV, there does not exist any close relation between the total hours of light received and the time of onset of maturity. It is probable that the duration of light at different phases of maturity influences the gonad differently. Even a reduced rate of light could not stop the maturation process; (Experiment No. 2), it only delayed the process a few days as compared to the control. In experiment No. 5 the delay is apparent only in the case of the female cycle. Males in that group oozed milt somewhat in advance of control. The results of experiments Nos. 6 and 7, where the fish matured before the normal season, in spite of receiving reduced light initially, is quite significant. Even at the time when the fish became mature the total hours of light received by them during the course of the experiment were much less. This leads one to presume that either the early phase of gonad development, in this fish, requires only reduced light or is more or less independent of the influence of light. If long photoperiods had any retarding effect in the early phase, fish under the 14 and 18 hours of light per day would have matured late. In the present stage of our knowledge it can be inferred with some degree of certainty that artificial day lengths longer than natural days accelerate maturation in the carp Cirrhina reba.

Early maturation and spawning of fish as a result of enhanced photoperiodic regimes have been reported in the case of Notropis bifrenatus (Harrington, 1950, 1957), Enneacanthus obesus (Harrington, 1956), Fundulus confluentus (Harrington, 1959a) and Orysias latipes (Yoshioka, 1963) among others. Bullough (1939) has observed that long photoperiods in combination with higher water temperature will hasten the development of ova in Phoxinus laevis. Contrary to these observations, the winter spawning fish of the northern latitudes like, Salvelinus fontinalis (Hoover and Hubbard, 1937; Hazard and Eddy, 1951; Corson, 1955 and Shiraishi and Fukuda, 1966), Onchorhynchus nerka (Combs et al., 1959; Shiraishi and Fukuda, 1966), Salmo gairdneri, Onchorhynchus rhodurus (Shiraishi and Fukuda, 1966) and Plecoglossus altivelis (Shiraishi and Takeda, 1961) are reported to have attained early maturity under experimental conditions of short light periods and delayed maturation under long light periods.

The above examples show that light has no uniform influence on the sexual development of fish. Hoover and Hubbard (1937), attempted to classify fish and mammals into three groups based on the breeding season. They are: (a) Those animals that breed as the days become shorter; Sheep, Deer, Brook trout and Land-locked Salmon, (b) Those which breed as the days become longer; Raccoons, some catfish and Black bass and (c) Those which breed without demonstrated relation to the length of day; Cats, Dogs and some aquaria fish. According to Yoshioka (1962), from the stand-point of the manner of response to light, two types of fish exist, long day type and short day type. Indian carps including *Cirrhina reba* breed naturally when the days become longer during the monsoon months. The experimental evidence collection on *C. reba* show that long light periods influence the reproductive cycle to induce early maturity and hence, it evidently falls under the "long day" type.

ACKNOWLEDGEMENT

The author is grateful to Dr. V.R. Pantulu, for suggesting the problem and initial guidance and to Dr. V.G. Jhingran, Director of the Institute and Dr. V. Gopalakrishnan for their encouragement and guidance throughout the work.

REFERENCES

- Alikunhi, K.H., M.A. Vijayalkshmanan and K.H. Ibrahim (1960). Preliminary Observations on the Spawning of Indian Carps, Induced by Injection of Pituitary Hormones. <u>Indian J. Fish</u>. 7: 1-19.
- Alikunhi, K.H., K.K. Sukumaran, S. Parameswaran and S.C. Banerjee (1964). Preliminary Observations on Commercial Breeding of Indian Carps Under Controlled Temperature in the Laboratory. <u>Central Inland Fish. Res.</u> Inst. Barrackpore Bull. 3: 20 pp.
- Allison, L. (1951). Delay in Spawning in Eastern Brook Trout by Means of Artificial Prolonged Light Intervals. <u>Procs. Fish. Cult</u>. 17: 111-116.
- Alm, G. (1959). Connection Between Maturity, Size and Age in Fishes. <u>Rep</u>. <u>Inst. Freshwater Res. Drottiningholm</u>. 40: 5-145.
- Atz, J.W. (1957). The Relation of the Pituitary to Reproduction in Fishes. <u>In: The Physiology of the Pituitary Gland of Fishes</u>. <u>New York Zoo-logical Soc</u>., New York.

- Bhimachar, B.S. and S.D. Tripathi (1967). A Review of Culture Fisheries Activities in India. <u>FAO Fish. Rep.</u> 44(2): 1-33.
- Bogenschutz, R.P. and H.P. Clemens (1967). Changes in the Pituitary Gland of Gold Fish *Carassius auratus*, during Diet Controlled Gonadal Regression. <u>Copeia</u>. 4: 827-835.
- Bullough, W.S. (1939). A Study of the Reproductive Cycle of the Minnow in Relation to the Environment. <u>Proc. Zool. Soc., London</u>. 109: 79-102.
- Chaudhuri, H. (1960). Experiments on Induced Spawning of Indian Carps with Pituitary Injections. <u>Indian J. Fish</u>. 7: 20-48.
- ----- (1963). Induced Spawning of Indian Carps. Proc. Wat. Inst. Sci. India. 29: 478-487.
- Clemens, H.P. and C.A. Reed (1967). Long-term Gonadal Growth and Maturation of Gold Fish (*Carassius auratus*) with Pituitary Injections. <u>Copeia</u>. (2): 465-466.
- Cobs, B.D., R.E. Burrows and R.G. Bigej (1959). The Effect of Controlled Light or the Maturation of Adult Blue Back Salmon. <u>Progr. Fish</u>. <u>Cult</u>. 21: 63-69.
- Carson, B.E. (1955). Four Years Progress in the Use of Artificially Controlled Light to Induce Early Spawning of Brook Trout. Progr. Fish. Cult. 17: 99-102.
- Harrington, J.W. (1950). Pre-seasonal Breeding by the Bridled Shiner. Notropis bifrinatus, Induced under Light-temperature Control. <u>Copeia</u>. (4): 304-311.
- ----- (1956). An Experiment on the Effects of Contrasting Daily Photo-periods on Gametogenesis and Reproduction in the Centrarchid Fish, Enneacanthus obesus (Girard). J. Exp. Zool. 131: 203-223.
- ------ Sexual Photoperiodicity of the Cyprinid Fish, Notropis brifinatus (Cope), in Relation to the Phases of Its Annual Reproductive Cycle. J. Exp. Zool. 135: 529-556.
- Daylength on the Ovogenetic Cycle of a Low-latitude Fish, Fundulus confluentus Goode and Bean. Zoologica. 44: 149-168.
- Annual Sexual Cycle. In: Photoperiodism in Fishes in Relation to the Plants and Animals, edited by R.B. Withrow. American Association for the Advancement of Science, Washington.
- Hazard, F.R. and R.E. Eddy (1951). Modification of the Sexual Cycle Brook Trout (Salvelinus fontinalis) by Control of Light. <u>Trans. Amer.</u> <u>Fish. Soc</u>. 80: 158-162.

- Henderson, N.E. (1963). Influence of Light and Temperature on the Reproductive Cycle of the Eastern Brook Trout, Salvelinus fontinalis (Michill). J. Fish. Res. Bd. Canada. 20: 859-897.
- Hoar, W.S. (1957). Endocrine Organs. In: <u>The Physiology of Fishes</u>, Edited by M.E. Brown, <u>New York Academy Press</u>, <u>New York</u>.
- Hoover, E.E. (1937). Experimental Modification of the Sexual Cycle of Trout by Control of Light. <u>Science</u>. 86: 425-426.
- Hoover, E.E. and H.E. Hubbard (1937). Modification of the Sexual Cycle of Trout by Control of Light. <u>Copeia</u>. 206-210.
- Pickford, G.E. and J.W. Atz (1957). The Physiology of Pituitary Gland of Fishes. <u>New York Zoological Society</u>, New York, p. 613.
- Shiraishi, Y. and Z. Fukuda (1966). The Relation between the Day-Length and the Maturation in Four Species of Salmonid Fish. <u>Bull. Freshwater</u> <u>Fish. Res. Lab. Tokyo</u>. 16: 103-111.
- Shiraishi, Y. and T. Takeda (1961). The Influence of Photoperiodicity on the Maturation of Ayu-fish, *Plecoglossus altivelis*. <u>Bull. Freshwater Fish</u>. <u>Res. Lab. Tokyo</u>. 11: 69-81.
- Verghese, P.U. (1967). Prolongation of Spawning Season in the Carp Cirrhina reba (Ham.) by Artificial Light Treatment. <u>Curr. Sci</u>. 36: 465-467.
- Yoshioka, H. (1962). On the Effects of Environmental Factors upon the Reproduction of Fishes. 1. The Effects of Day-length on the Reproduction of the Japanese Killifish, Oryzias latipes. <u>Bull. Fac. Fish.</u> <u>Hokkaido Univ. 13: 123-136.</u>
- production of Fishes. <u>Bull. Fac. Fish. Hokkaido Univ.</u> 14: 137-151.

Table I

Details of light conditions employed for experiments 1 - 7

Months	Control Ex.No.1 Nat.day	rol o.l day le	Control Ex. No.2 Ex.No.1 Retarded Nat.day length [*] period. ⁸	Ex. No.2 Retarded period. {	lo.2 ded ph d.8 h	Photo- Ex.No. 3 Ex.No. 4 Photo- Long photo- Long photo- 8 hrs/day period.18hr/day	Ex.No. Long pl period	Ex.No. 3 Long photo- period.l 4 hr	- r/day	Ex.No. Long pl period	Ex.No. 4 Long photo- period.18hr	- ^/day	Ex. No. 5 Varying photo- period.20-8- 18hr/day	o. 5 ng ph d.20-	oto- 8-	Ex. Nos. 6 & 7 Varying photo- period. 4-14-2 hr/dav	5.6 18 pho 1.4-1	6 & 7 photo- 4-14-20
	d.a.	n T	d.a. m.t. c.t.	d.a.	m.t.	c.t.	d.a.	m.t.	c.t.	d.a.	n t	c.t.	d.a. m.t.		c.t	d.a.	m.t.	c.t.
August 1966	12.91	004	004	8 hr	248	248	14	434	434	18 1	558	558 1	18.13	562	562	5.87	182	182
Sept. "	12.29	368	768	=	240	488	=	420	854	E	540 1	1098 1	13.80	414	976 1	10.20	306	488
October "	11.64	361	1129	:		736	=	434	1288	=	558]	1656	9.50	294 I	1270 1	13.74	426	914
November "	11.21	336	1465	:	240	976	5	420	1708		540 2	2196	8.00	240 1	1510 1	14,00	420	1334
December "	10.78	334	1799	=	248	1224	Ξ	#34	2142	=	558 2	2754	=	248 L	1758	=	434	1768
Jan. 1967	10.91	338	2137	-	248	1472	=	H34	2576	=	558 3	3312	=	248 2	2006	=	434	2202
Feb. "	11.39	319	2456	=	224	1696	:	392	2968	E	504 3	3816 P	9.25	259 2	2265 1	5.25	427	2629
March "	12.00		2828	=	248	1944		434	3402	Ξ	558 4	4374]	11.26	349 2	2614 1	7.28	535	3164
April "	12.65	379	3207	=	240	2184	-	420	3822	E	540 1	1914 1	13.30	399 3	3013 1	19.60	578	3742
May "	13.20		3616	=	248	2432	*	434	4256	E	558 5	5472]	15.26 4	473 3	3486 2	20.00	620	4362
June "	13.50	405	H021	=	240	2672	-	420	46.76	-	540 5	6012 J	7.26	518 4	4004 2	20.00	600	4962
July "	13.37	414	4435	1	I	I	1	1		1	I	1	18.00 S	558 4	4562	l	I	I
								***					 			,		

d.a. - Average duration of light per day
m.t. - Total hours of light received during the month
c.t. - Cumulative total hours of light received from the initiation of the experiment.

st Natural day length is calculated from the table for sun rise and sun set at Calcutta.

Table II

	Average ma:	kimum water temperatu	ure in ^o C
Month	Control Ex. No. 1	Experiments Nos. 2, 3, 4, 5 and 6	Experiment No. 7
August 1966	28.8	29.1	28.9
September "	27.4	27.7	31.0
October "	26.1	26.5	30.2
November "	23.8	24.0	27.1
December "	19.6	20.1	27.1
January 1967	19.4	19.5	28.5
February "	21.8	22.5	29.5
March "	24,5	25.0	29.7
April "	27.0	27.5	29.9
May ''	29.7	29.8	29.8
June "	30.0	30.2	

Average water temperature in the cisterns during experimental period

Table III

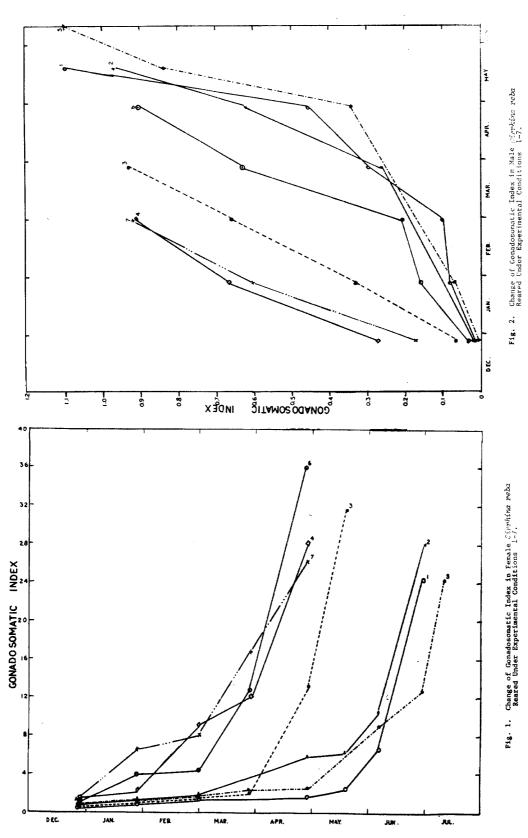
Experiment		iculars of fish at xperimental period	
Number	Total No. of fish	Average Total- length in mm.	Range of T.L. in mm.
1	40	84,70	63-99
2	34	82.62	76-100
3	33	83.09	69-103
4	43	80.46	65 -100
5	43	83.05	70-100
6	46	81.11	70-102
7	47	80.64	64-101

Number and size of Cirrhina reba used for the experiment

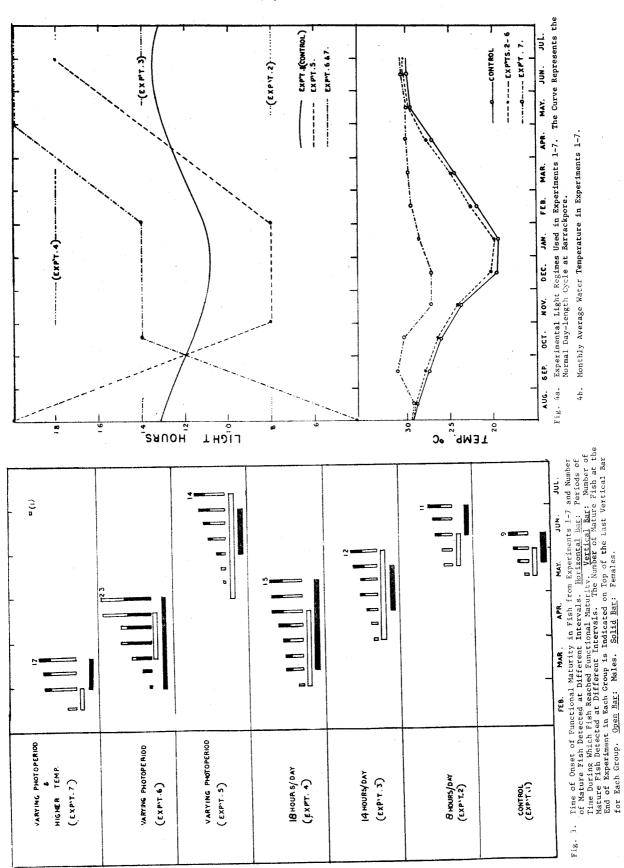
Maturity retarded or advanced	compared to control	Norma 1		M. retarded by 10 days	ret	by 21 days M. 41 days	advance	in advance		ln advance F. 71 davs			ın advance F. retarded	51	adi	F. 81 days in advance		in advance		in advance
Size range of mature fish T.L. in mm.	Female	128	150	103	136	108	ן סכי	C 7 T	06 ,	т 125	!	98 ,	۲ 121	 82	н , ,	128	86	Ч	121	
Size matu T.L.	Male	132	162	105	134	104	ц г) 7	96	т 126		100	130	100	ч,	L32	86	Ч	119	
hours of given till maturity	Female	3468)	2504		3668			3978			3470		2629			2629			
Total light first		3326		2300		3374			3816			3013		2950			2408			
on which function- mature fish first recorded	female	20.5.67		10.6.67		20.4.67			10.3.67			30.5.67		28.2.67			28.2.67			
Date on which ally mature f recorded	oozing male	10.5.67		20.5.67		30.3.67	662 (28.2.67			30.4.67		20.3.67			15.2.67			
Temperature range degrees C		28.8-19.4-30.0		29.1-19.5-30.2	-	- do -			- do -			- do -		- do -			28.1-27.1-30.2			
Experiment Nature of light Number period		Control natural		Artificial light 8 hr. per day		Artificial light	14 hr. per day	Artificial light	18 hr. per day			Artificial light-	varyıнg ∠∪/о/тонг /day	Artificial light-	Varying 4/14/20	Apn / TH	As above			
Experiment Number		-		0		ო		#				ى		Q			7			

Summary of the experiments

Table IV



- 183 -



- 184 -