THE FEMALE REPRODUCTIVE CYCLE OF THE CRAYFISH *CAMBARELLUS SHUFELDTI:* THE INFLUENCE OF ENVIRONMENTAL FACTORS[1]

MILDRED EILEEN LOWE,

Department of Zoology, University of Melbourne, Melbourne, Australia

Many organisms show a reproductive periodicity correlated with environmental factors. Photoperiodism and response to changing temperature are the best known phenomena. Food and rainfall and, in marine organisms, changes in salinity and changes in pressure due to spring tides, may also affect time of egg-laying.

Most of the research on influence of photoperiodism upon reproductive cycles in invertebrates concerns induction of diapause in insects. This subject was reviewed by Lees (1955). Most insects may be classified as "long-day" species; that is, exposure to long day-lengths allows uninterrupted development while short day-lengths favor diapause. However, in many insects extremely short day-lengths or constant darkness may also prevent diapause.

High temperatures augment the effect of long day-lengths in preventing diapause, while low temperatures favor its onset. For the termination of diapause, temperature may be more important than day-length as many insects require a period of chilling before diapause terminates (Lees, 1955).

By increasing the temperature in midwinter Loosanoff and Davis (1952) initiated a second breeding season in the oyster *Crassostrea virginica* which normally breeds only during the summer. Low temperature stimulated breeding in *Balanus*, while spawning occurred both at time of maximum and of minimum temperatures in the limpet *Patella vulgata* (Giese, 1959).

Korringa (1952) showed that temperature is not the only factor influencing spawning. Within the spawning season of the European oyster, *Ostrea edulis*, semimonthly peaks of reproductive activity are correlated with spring tides. In this and other animals exhibiting increased activity two times each lunar cycle, the increased water pressure of spring tides might be the triggering influence. Monthly spawning, as in the palolo worm, is believed to be iniated by increased photoperiod (reviewed by Korringa, 1947).

Among the Crustacea, G. J. Stephens (1952) showed, in *Orconectes virilis*, that increased day-length accelerated the rate of the normal cyclic increase and decrease in ovarian size. She also demonstrated that constant darkness completely interrupted cyclic activity, the oocytes developing to a mature condition and remaining in that state. However, Suko (1958) showed that the response of *Procambarus clarkii* to darkness depends on the state of the ovary when the animal is placed in the darkness. With another crayfish, *Orconectes rusticus*, G. C. Stephens (1952) showed that light caused modifications of a secondary sexual structure, the cement glands, which secrete a substance used in attaching the newly-laid eggs to the pleopods. These glands were stimulated to develop at a more rapid rate in animals under increased photoperiods.

The present investigation was undertaken to define the female reproductive cycle of the crayfish *Cambarellus shufeldti*. This small crayfish is readily available in southeastern Louisiana throughout the year. It has been used in studies on chromatophores, retinal pigment migration, metabolism, and on the endocrine factors controlling these functions (*e.g.*, Fingerman, 1957; Fingerman and Lowe, 1957). The only studies of the life cycle of this species were the brief ecological surveys of Penn (1942, 1950) who reported an almost continual period of reproduction with peak activity in late winter, continuing high through early summer.

The series of observations presented here represents an attempt (1) to outline in detail the female reproductive cycle of this crayfish, and (2) to obtain information concerning environmental factors that may regulate the cycle.

---

[1] Most of this paper is a portion of a dissertation submitted in partial fulfilment of the requirements for the Ph.D. degree in zoology at Tulane University, New Orleans, Louisiana, 1959.

Materials and Methods

Adult specimens of *Cambarellus shufeldti* are small, the female averaging only 20 mm in length. Typically they inhabit shallow fresh-water ponds and ditches that are exposed to direct sunlight part of each day and that contain an abundant plant growth (Penn, 1942, 1950). The population studied occurred in a roadside ditch, in pinelands, near Pearl River, Louisiana. The principal water plants present were *Scirpus nanus* and *Rhyncospora* spp. Other crayfishes associated with *Cambarellus* were *Orconectes clypeatus* and *Procambarus blandingi acutus.*

The water level in the ditch fluctuated from approximately 18 inches to below ground level according to the amount of rainfall. Very little rain fell during late autumn of 1958 and the ditch was dry from the middle of December to the third week in January, 1959. The crayfish can survive such droughts by burrowing. They evidently do not burrow unless there is no standing water as a small December collection was obtained from only one-half inch of water by raking through plant growth. At all other periods during the study the crayfish were active and were obtained with dipnets. Although a thin film of ice formed over the water surface in February, 1958, animals were still present in the water.

Animals were collected at least once a month from November, 1957, through November, 1959, preserved in Bouin's fixative, and stored in 70 percent ethyl alcohol. The weights and measurements presented below are those obtained from preserved crayfish. The animals were blotted and weighed to the nearest milligram. Cephalothorax length, from tip of rostrum to posterior margin of carapace, was measured to the nearest 0.5 mm with the aid of dividers and a stereoscopic microscope.

The degree of cement gland development and the presence or absence of a plug in the oviducts were noted. In *C. shufeldti* the cement glands underlie the anterior portions of the second through sixth abdominal segments at the junction of the sterna and pleura and the mid-ventral line along the strong transverse bar present in each sternum. The glands extend into the protopodite and endopodite of each pleopod and into both the exopodite and endopodite of the uropods. The degree of cement gland development was determined according to the stages of G. C. Stephens (1952): stage 1. Numerous tiny, milky-white translucent, circular or subcircular spots appear in areas of future gland development; stage 2. The white areas enlarge and subcircular transparent areas appear within them; stage 3. The glands appear as translucent white lobate clusters surrounding the original white areas in an irregular manner; stage 4. The glands become opaque, milky-white and so filled with secretion that their lobate character is difficult to distinguish.

In *Cambarellus* the mid-ventral glands mature slowly and may still be in stage 2 or 3 when the lateral glands are in the fully developed stage 4. Therefore, the states of the two sets of glands were recorded separately. A stage listed as 4.0, 2.0 would indicate that the lateral glands were at stage 4, the mid-ventral only at stage 2.

After the specimens had been examined grossly their ovaries were removed. The ovary is Y-shaped with two anterior and one posterior lobes located immediately ventral to the heart; the anterior lobes curve dorsally around its anterior end. The oviducts originate from the ventro-lateral surface of the ovary where the three lobes join. Camera lucida drawings were made of the ovaries before removal from the animal, with the crayfish held in a lateral and slightly dorsal position.

The diameters of the largest oocytes in each ovary were measured to the nearest 0.01 mm with an ocular micrometer and the average diameter of the four largest was recorded. Each ovary was weighed to the nearest 0.1 mg. after being touched to a piece of filter paper and placed in a dry, covered weighing bottle. The weight of any moisture remaining in the weighing bottle when the ovary was removed was subtracted from the previously recorded weight.

Results and Discussion

The Normal Yearly Reproductive Cycle

Animals were collected at least once a month from November, 1957, through November, 1959, to determine the normal reproductive cycle. Only animals of 8.5 mm cephalothorax length and larger were considered because only two mature animals smaller than this were found.

A population of *C. shufeldti* has an almost continuous period of reproductive activity. Females bearing eggs or young can be collected nearly every month of the year. The eggs are attached to the female's pleopods, where young hatch from the eggs and remain attached through the second instar. The total time of attachment to the pleopods in *Cambarellus* is about three weeks.

The percentage of females bearing eggs and young is shown in Figure 1. Two peaks of reproductive activity occur, the higher in late winter and a second in early summer. A small rise also occurs in October.

The life cycle.—Approximately fifty females from each of the monthly collections from January, 1958, through November, 1959, were weighed and cephalothorax measured. Animals carrying eggs or young were weighed with brood attached. Each month some animals were dissected to determine ovarian condition.

Comparison of total body weight (Tables 1 and 2) shows three distinct groups of females which appear and mature at different times during the year. There is a definite separation between the heaviest animals in one group and the lightest in a second, heavier group present in the population at the same time. Comparison of increment in cephalothorax length produces only a hazy pattern of growth as seen in Table 2 which summarizes the correspondence between weight and length throughout the study.

In January 1958 only one size group of adult females was present. The same group (hereafter referred to as group A) was present in February, showing a small increment in weight and no increase in length. Eighty-five percent of these animals carried eggs or young. The largest juveniles measured 7.5 mm cephalothorax length; they were not weighed.

By March many group A animals had lost their young and molted, and the group showed a large weight increase. A second group of females (group B) had molted to mature size. These latter comprised 36 percent of the adult female population and all were ovigerous. The largest juvenile measured 7.5 mm cephalothorax length.

By April 1958 all remaining group B animals had undergone a maturation molt as the largest juveniles measured only 6.0 mm cephalothorax length. Approximately 50 percent of the group carried eggs or young. All of group A had completed a post breeding molt. These two groups could not be separated on the basis of length as group A measured 9.0-10.5 mm cephalothorax length and group B, 10.0-11.5 mm.

TABLE 1.
Body weight increment in the three groups throughout the study.

Month	Group A	Group B	Group C
1-58	164-206 (190)*		
2-58	190-223 (203)		
3-58	256-310 (284)	181-200 (190)	
4-58	292-387 (340)	186-215 (192)	
5-58	315	193-240 (215)	
6-58		268-340 (299)	196-227 (212)
7-58		329-359 (341)	148-265 (197)
8-58			163-253 (199)
9-58			196-245 (221)
10-58			235-384 (274)
11-58	158-185 (172)		292-364 (311)
12-58	178-210 (196)		300-345 (327)
1-59	192-230 (210)		377-418 (393)
2-59	215-292 (251)		
3-59	256-310 (284)	167-201 (185)	
4-59	305-378 (337)	179-220 (200)	
5-59		181-250 (220)	
6-59		234-290 (278)	150-162 (156)
7-59		287-335 (310)	174-218 (194)
8-59		320-335 (330)	193-227 (211)
9-59			208-231 (218)
10-59			245-286 (265)
11-59	160-188 (175)		279-317 (299)

* 164-206 = weight range; (190) = mean.

TABLE 2.
Monthly weights and cephalothorax lengths of a representative sample of the adult female population.

Month	Group	No. Measured & Weighed	Cephalothorax Length (mm) range	mean	Weight (mg) range	mean	% in size class
Jan. 1958	A	35	9.0-10.0	9.6	164-206	190	100
Feb. 1958	A	41	9.0-10.5	9.8	190-223	203	100
March 1958	A	36	9.5-11.0	10.0	256-310	284	64
	B	20	9.0- 9.5	9.2	181-200	190	36
April 1958	A	19	10.0-11.5	10.5	292-387	340	40
	B	28	9.0-10.5	9.7	186-215	192	60
May 1958	A	1	10.5		315		2
	B	42	9.0-10.5	9.8	193-240	215	98
June 1958	B	46	9.0-11.5	10.1	268-340	299	76
	C	15	9.0-10.0*	9.4	196-227	212	24
July 1958	B	6	10.5-11.5	11.0	329-359	341	12
	C	44	8.5-10.0	9.2	148-265	197	88
Aug. 1958	C	50	8.5-10.0	9.2	163-253	199	100
Sept. 1958	C	51	9.0-10.5	9.6	196-245	221	100
Oct. 1958	C	50	9.5-11.5	10.0	235-384	274	100
Nov. 1958	C	52	9.5-11.5	10.2	292-364	311	80
	A	13	9.0- 9.5*	9.2	158-185	172	20
Dec. 1958	C	9	10.0-11.5	10.6	300-345	327	30
	A	21	9.0-10.0	9.6	178-210	196	70
Jan. 1959	C	4	10.0-11.5	11.0	377-418	393	9
	A	45	9.5-10.5	10.0	192-230	210	91
Feb. 1959	A	45	9.5-10.0	9.9	215-292	251	100
March 1959	A	39	9.5-11.0	10.1	265-315	288	78
	B	11	9.0- 9.5	9.2	167-201	185	22
April 1959	A	24	10.0-11.5	10.6	305-378	337	44
	B	31	9.0-10.0	9.6	179-220	200	56
May 1959	B	35	*9.0-10.5	9.7	181-250	220	100
June 1959	B	48	9.5-11.0	10.1	234-290	278	92
	C	4	*9.0-10.0	9.3	150-162	156	8
July 1959	B	14	10.5-11.5	10.8	287-335	310	28
	C	36	8.5-10.0	9.2	174-218	194	72
Aug. 1959	B	3	10.5-11.5	11.0	320-335	330	6
	C	47	8.0-10.5	9.5	193-227	211	94
Sept. 1959	C	35	8.5-11.0	9.9	208-231	218	100
Oct. 1959	C	35	9.5-10.5	10.2	245-286	265	100
Nov. 1959	C	48	10.0-11.5	10.6	279-327	299	85
	A	9	8.5- 9.5	9.0	160-188	175	15

* In these groups 8.5 mm animals were present but are not included in the tabulation as their ovaries were in an immature condition.

However, animals measuring 10.0 and 10.5 mm in group A were approximately 100 mg heavier than those of the same length in group B.

Group A comprised only 2 percent of the population by May, 1958. Most group B individuals had lost the young from the pleopods but had not undergone a post-breeding molt.

By June most of group B had molted and 65 percent of these had produced a second brood. A third group of females, group C, had begun to mature and formed 2 percent of the adult female population. Approximately 85 percent of this new group were ovigerous. The ovaries of 8.5 mm females that now appeared in the population were immature.

In July the remainder of group B molted and all the animals were large, 10.5 to 11.5 mm cephalothorax length. This group comprised 12 percent of the population and none was ovigerous. More group C animals had undergone a maturation molt and the

ovaries of 8.5 mm animals reached a mature condition. Group C could be subdivided into two sections, the larger representing those animals hatched at the beginning of the winter breeding season and reproducing in June, the smaller those hatched towards the end of the season and reproducing in July and August.

By August 1958 group B had disappeared from the population. The 18 percent of group C that carried eggs or young was composed of smaller animals, 8.5 to 9.0 mm. There were many juveniles whose cephalothorax length was 8.0 mm. The larger animals had shown no increase in weight.

In September 1958 a number of the animals molted as shown by increase in length, but the weight of the larger animals remained stationary.

By October all the animals molted and there was a large weight increment. The largest animals produced a second brood.

In November the larger animals of group C were dying off without molting again. The group made up 86 percent of the adult females as Group A began to appear in the adult population. The 8.5 mm animals possessed immature ovaries. Only in July and August were such small animals found in a reproductive phase.

In December only a small collection could be obtained due to drought conditions. Group C constituted 30 percent of the adult female population. Some Group A animals had molted but the increment in weight was small.

In late January, 1959, group C was composed of very large, heavy animals, all of which were ovigerous. Group A animals molted but had not yet laid eggs.

By February the entire adult female population was again composed of only one size group, group A, 63 percent of which were carrying eggs or young.

Figure 1 and Tables 1 and 2 show the repetition of this cycle through the rest of 1959.

Maturation of oocytes.—In the ovaries of immature animals the oocytes are tiny, 0.40 mm or less in diameter. They are transparent and widely separated by interstitial material. As the animals mature the oocytes begin to grow, become translucent white and then

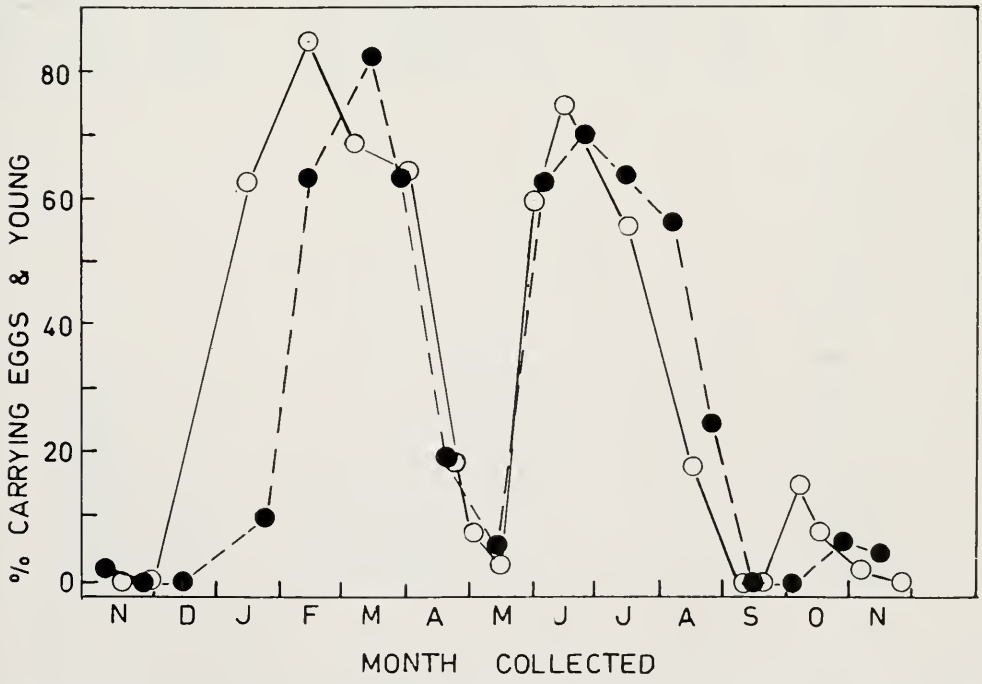

Figure 1. The yearly reproductive cycle of *C. shufeldti* expressed in terms of the percentage of females that were carrying eggs or young. Solid line, circles = Nov. 1957-Nov. 1958; dotted line, dots = Nov. 1958 - Nov. 1959.

opaque ivory at approximately 0.70 mm in diameter. The oocytes later acquire a greenish tinge as yolk deposition increases their size to 1.1 mm. The color of the oocytes gradually deepens to the dark dull green-black of mature eggs, with a diameter of 1.4 to 2.0 mm. Eggs are black immediately after laying. The smallest eggs found attached to the pleopods measured 1.45 mm. The same sequence of oocyte development is repeated after egg-laying occurs.

At times yolk resorption occurs and the oocytes may become a bright chicken-yolk yellow or even orange. Oocytes undergoing resorption become coarsely granular, irregular vacuoles appear within them, and they become surrounded by a clear yellow fluid. When resorption is occurring rapidly the entire ovary may become distended with a pale yellow fluid. As resorption proceeds the oocytes become smaller and their outlines irregular. They eventually disinte-

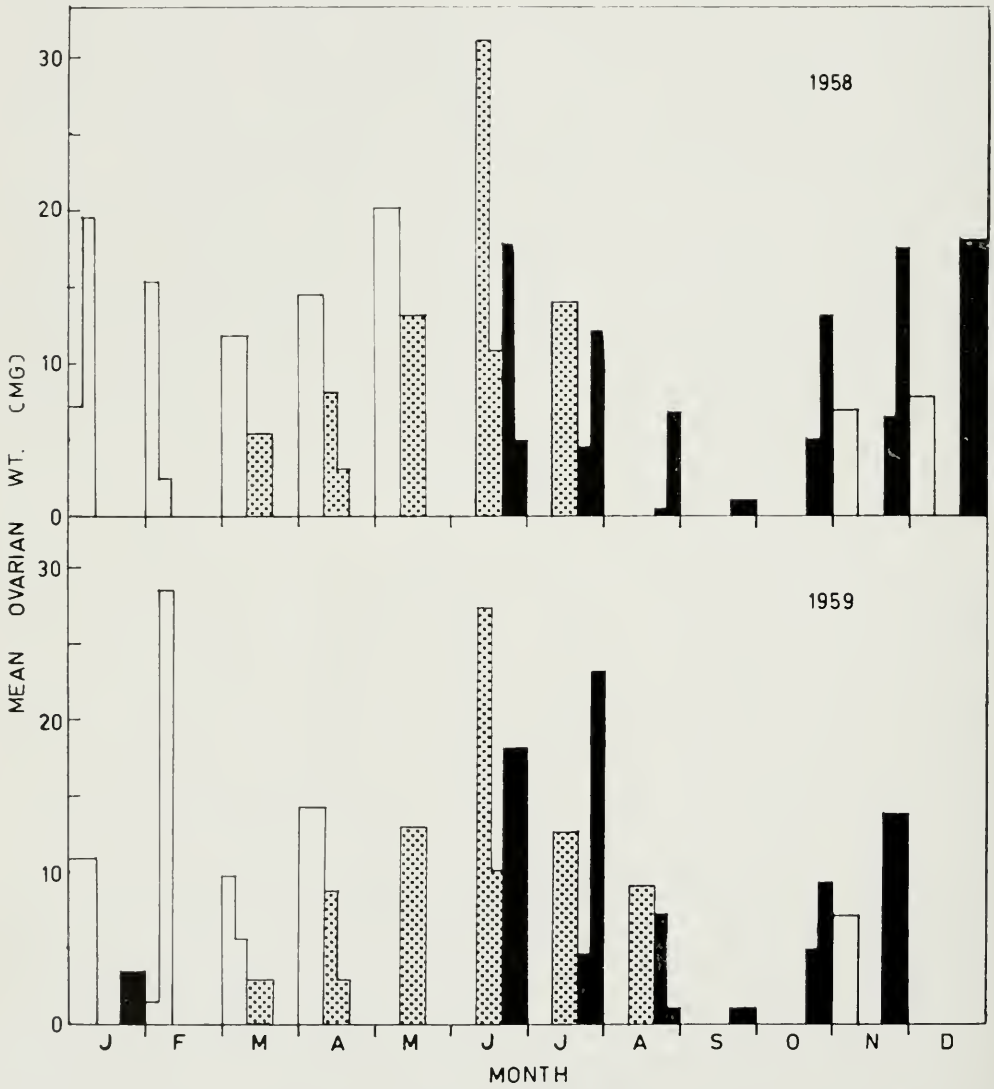

Figure 2. Variation in mean ovarian weight throughout the year. Group A, open; Group B stippled; Group C solid bars. When two values are given for the same group in one month, the lesser value refers to animals carrying eggs or young at time of measurement.

grate leaving loose irregular masses of yellow-orange granules that gradually disappear.

Ovarian cycle. — The condition of the ovaries reflects the presence of the three size groups. Thus the ovarian weights of the

TABLE 3.
Summary of monthly measurements on ovarian weight and oocyte diameter.

Month	Group	No. used	Weight of Ovary (mg) range	mean	Oocyte Diameter (mm) range	mean
1-58	A*	14	5.5- 8.2	7.2	1.17-1.33	1.25
	A	12	18.7-20.3	19.5	1.63-2.00	1.83
2-58	A-	13	13.2-17.3	15.3	1.33-1.57	1.51
	A*	10	1.7- 2.7	2.4	0.37-0.67	0.49
3-58	A*-	12	10.9-19.7	11.8	0.92-1.16	0.98
	B*	15	5.2- 5.6	5.4	0.58-0.68	0.64
4-58	A-	14	13.9-15.6	14.6	0.85-0.98	0.92
	A*	1	9.2		0.83	
	B-	13	7.4- 8.9	8.1	0.69-0.98	0.89
	B*	10	2.5- 3.7	3.1	0.58-0.67	0.63
5-58	A-	1	21.0		1.25	
	B	11	9.1-16.9	13.2	0.84-1.18	1.04
6-58	B	10	30.9-32.1	31.5	1.67	1.67
	B*-	14	9.8-12.0	10.7	0.84-1.08	0.98
	C	7	16.2-19.5	17.8	1.34-1.68	1.50
	C*	8	3.8- 5.8	4.9	0.66-0.92	0.81
7-58	B-	4	13.7-14.5	14.0	1.46-1.51	1.48
	C-	13	10.2-14.4	12.1	1.35-1.53	1.41
	C*	10	3.1- 5.8	4.5	0.50-0.82	0.68
8-58	C	10	6.0- 7.2	6.7	1.10-1.18	1.15
	C*	9	0.4- 0.7	0.5	0.38-0.50	0.41
9-58	C	13	0.7- 1.5	1.1	0.35-0.66	0.44
10-58	C	11	11.5-15.1	13.2	1.25-1.43	1.34
	C*	9	4.1- 6.5	5.0	0.75-0.83	0.79
11-58	C	14	16.0-18.7	17.5	1.25-1.43	1.35
	C-	11	6.0- 7.0	6.5	0.78-0.92	0.85
	A	13	6.1- 7.8	7.0	1.23-1.33	1.28
12-58	C	9	17.0-19.4	18.1	1.48-1.60	1.53
	A	10	7.2- 8.4	7.8	1.26-1.34	1.30
1-59	C*	9	3.4- 3.8	3.6	0.80-0.85	0.83
	A	12	9.3-12.8	10.9	1.33-1.67	1.51
2-59	A*	11	1.2- 1.8	1.5	0.37-0.50	0.43
	A	10	28.0-29.3	28.6	1.75-2.00	1.85
3-59	A*	10	4.8- 6.7	5.6	0.65-0.83	0.74
	A-	12	7.6-11.5	9.5	0.87-1.14	0.96
	B*	10	2.0- 3.5	2.8	0.58-0.71	0.65
4-59	A-	8	12.8-16.6	14.2	0.84-0.99	0.91
	B*	11	2.1- 3.1	2.7	0.57-0.67	0.64
	B-	10	6.9- 9.8	8.5	0.75-0.96	0.87
5-59	B	13	8.5-16.3	12.8	0.87-1.20	1.05
6-59	B	12	25.1-30.8	27.4	1.63-1.68	1.65
	B*-	12	6.2-11.4	10.0	0.79-1.04	0.91
	C	4	16.4-20.0	18.1	1.40-1.61	1.51
7-59	B-	5	10.8-14.0	12.6	1.40-1.47	1.44
	C	10	21.3-28.1	23.2	1.51-1.73	1.63
	C*	12	3.0- 5.5	4.6	0.47-0.79	0.64
8-59	C & C-	8	6.7- 8.0	7.3	0.98-1.18	1.04
	C*	10	0.5- 2.0	1.2	0.43-0.58	0.51
	B-	5	8.0-10.1	9.0	1.20-1.31	1.24
9-59	C	12	0.7- 1.6	1.1	0.37-0.61	0.48
10-59	C	12	8.0-10.9	9.3	1.15-1.23	1.18
	C*	2	4.6- 5.0	4.8	0.68-0.74	0.71
11-59	C	12	12.8-15.8	13.8	1.20-1.38	1.28
	A	9	6.7- 8.0	7.2	1.16-1.31	1.23

* carrying eggs or young
- had carried young recently, signs of egg cases still on pleopods

three groups are plotted separately in Figure 2. However, in any one month the actual state of the ovaries of all non-ovigerous females is rather similar regardless of group, the heavier animals having heavier ovaries mainly because the ovary contains a larger number of eggs rather than eggs in a more advanced state of maturation. Table 3 which lists oocyte diameter and ovarian weight by group shows this clearly (see April, November). In most cases a single description in any one month will define ovarian condition in all non-ovigerous or in all ovigerous animals at that time. Figures 2 and 3 illustrate the sequence of ovarian changes.

January 1958. The ovaries of non-ovigerous females (Fig. 3, A) were large, the oocytes mature. The posterior lobe of the ovary was almost three times the length of each anterior lobe. The anterior lobes were rounded and thick. They curved backward dorsally over the heart and pressed against the carapace. The ovary extended from the bases of the first pereiopods to the bases of the second pleopods. The ovary of breeding females averaged 7.2 mg and was growing (Table 3).

February 1958. The ovaries of breeding

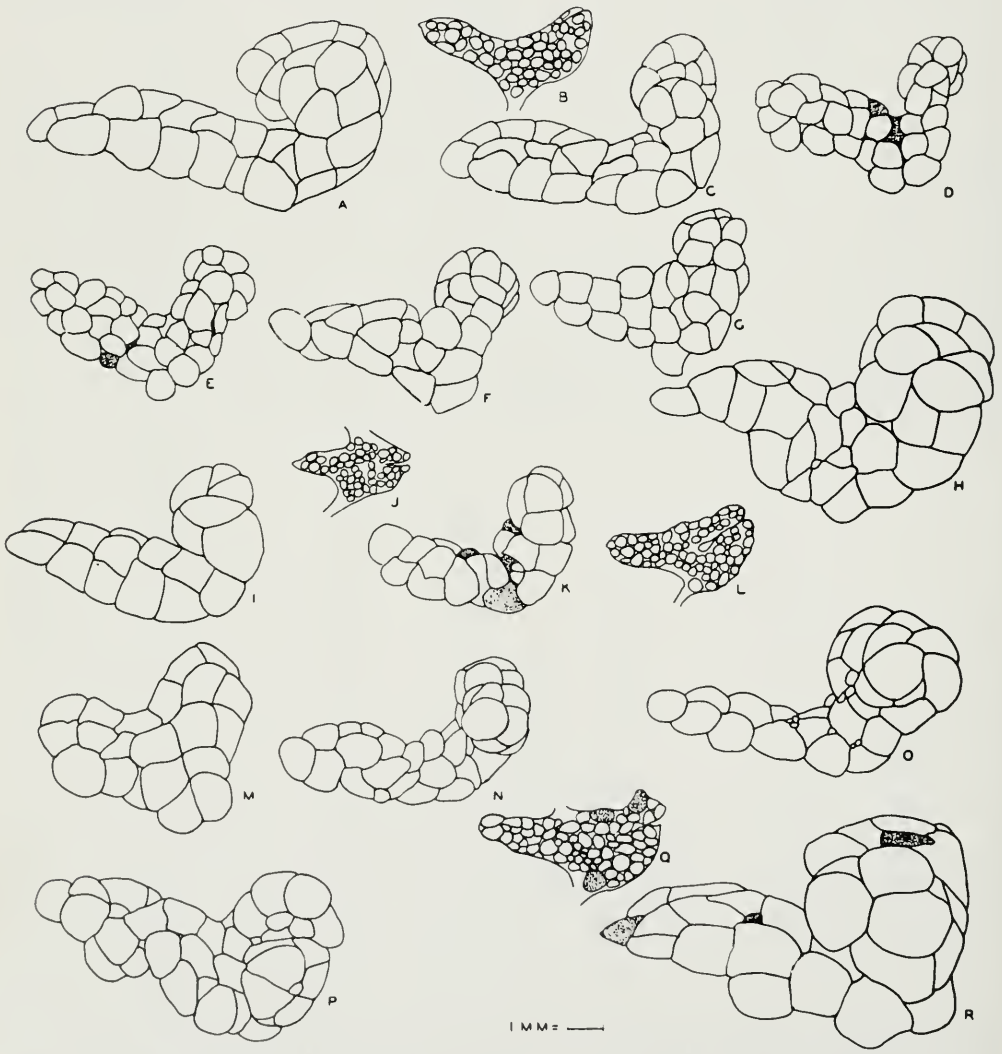

Figure 3. Annual cycle of ovarian development, January 1958 (3A) through February 1959 (3Q, R). See text for full explanation.

females were quite small and filled with new oocytes (Fig. 3, C). Those animals not carrying a brood retained egg cases; young had recently been present. The ovaries of this group had doubled their weight (Table 3) and retained the same general shape as present in January (Fig. 3, B).

March 1958. Group B had appeared and all were ovigerous. The animals of group A carried young or had molted after having a brood. About 5 percent of the oocytes present in recently molted animals showed yolk resorption, accounting for the decrease in ovarian weight. The posterior lobe of the ovary was more than twice the length of each anterior lobe (Fig. 3, D). The anterior lobes were elongated and narrow rather than rounded and thick as in January and February. The ovary extended from the second pereiopods just to the first abdominal segment.

April 1958. The animals carrying eggs had small ovaries, filled with new oocytes. The ovaries of the rest of the population had increased in size (Fig, 3, E) and were of the same shape as in March. Within an ovary the oocytes were not as uniform in size as previously; new oocytes were forming among more mature ones.

May 1958. Ovarian weight and oocyte diameter had increased (Fig. 3, F). The posterior lobe was approximately twice the length of each anterior lobe. Only 5 percent were brooding.

June 1958. In animals carrying broods the ovary (Fig. 3, G) was larger as contrasted with comparable groups in February, March and April, indicating a more rapid maturation of oocytes after egg-laying. In both ovigerous and non-ovigerous females the ovary was thick and compact, the posterior lobe only one and one-half times the length of each anterior lobe.

July 1958. Many of the non-ovigerous females still retained egg cases on their pleopods. The anterior lobes of the ovary were rounded and less than one-half the length of the posterior lobe (Fig. 3, I). The oocytes were larger than in June but had not increased in number.

August 1958. The ovaries of brooding animals were tiny, weighing only 0.5 mg and containing no oocytes larger than 0.42 mm (Fig. 3, J). This condition contrasts with that occurring in specimens collected in June when the ovary immediately began to increase in size and weight after egg-laying. The ovaries of those animals not carrying eggs or young averaged only 6.7 mg in weight and the oocytes were not uniform but of various sizes (Fig. 3, K). Yolk resorption was occurring; the ovary was reducing in size.

September 1958. No females carried eggs or young. The ovary was small, 1.1 mg, and extended from the bases of the third to the fifth pereiopods. The oocytes were round and of fairly uniform size; they were loosely arranged and one layer thick (Fig. 3, L), approximating the condition seen in immature animals.

October 1958. Fifteen per cent of the females carried eggs or young. The ovary of these animals weighed 5 mg and was growing. In those animals not carrying a brood the ovary averaged 13 mg in weight and oocytes measured 1.33 mm in diameter (Table 3). The ovary was thick and compact, extended from the second pereiopods to the end of the thorax. The three lobes were of about equal size. The anterior ones were upright with no backward curl (Fig. 3, M).

November 1958. Group A appeared in the population. The oocytes in this group underwent a slow maturation from November to February when there was a sudden increase in ovarian weight. In all females present in November the posterior lobe of the ovary was almost twice the length of the anterior lobes; the anterior lobes were beginning to enlarge laterally (Fig. 3, N).

December 1958. The ovary showed only a slight increase in weight over November. The posterior lobe had elongated to three times the length of the anterior ones. A few young oocytes were apparent (Fig. 3, O).

January 1959. Ten percent of the females were ovigerous. They represented the remnant of group C. The ovaries of the remaining 90 percent had the same general shape and extent as in December. While yolk deposition was occurring in the younger oocytes, resorption of yolk was beginning in some of the mature ones (Fig. 3, P).

February 1959. In brooding females the ovary was small and thin, but compactly arranged with little interstitial material (Fig. 3, Q). The largest oocytes measured 0.5 mm in diameter and represented old oocytes

from which yolk was being resorbed. Smaller, new oocytes were being proliferated. In those females which had not yet laid eggs the ovaries approximated the condition found in January 1958, but was heavier. Yolk resorption was occurring in about 10 percent of the oocytes (Fig. 3, R). This resorption was probably the result of forced retention of eggs past their maturation due to drought conditions at the end of 1958.

Cement gland development. — Cement gland development parallels oocyte maturation. The glands are not apparent in immature individuals. As the ovary approaches breeding condition the cement glands approach their fullest development. They decrease in size when egg-laying occurs and their secretion presumably provides the means of attaching eggs to the pleopods. After egg-laying the glands again begin to fill with secretion. Just as the ovary shows regression in late summer, so too do the cement glands, reducing to a stage of approximately 0.5, 0.0 in September from stage 4.0, 3.5 in July (Table 4). The sequence tabulated for 1958 also occurred during 1959.

The presence or absence of an externally viewed plug in the oviducts is also recorded in Table 4. The significance of this plug is not known. It appears to be composed of an amorphous or granular white secretion, probably from the ovary itself. It is not present in immature animals or in mature animals in September, but is present to some extent throughout the rest of the year, becoming very prominent and causing the thin exoskeleton covering the oviduct openings to bulge outward prior to egg-laying. Even when not apparent externally this white material may be found in the upper part of the oviduct. It appears to move down the oviduct and before egg laying is present only in the lower part of the duct. The substance is particularly prominent in the upper part of the oviduct and even in the ovary itself, near the duct, when yolk resorption is occurring.

Discussion. — The life span of female *C. shufeldti* is no longer than one year; that of an adult female, approximately six months, during which time she may produce two broods. Those animals which hatch in the late winter-early spring breed-

TABLE 4.
Average monthly state of cement gland development (fifty specimens were examined each month).

Month	Group A	Group B	Group C
January 1958	2.0, 0.0, slight*– 4.0, 3.0, yes		
February 1958	3.5, 1.0, yes 2.0, 0.0, yes–		
March 1958	3.5, 2.0, yes	1.5, 0.0, slight–	
April 1958	4.0, 2.0, yes	3.0, 2.0, yes 2.0, 0.0, yes–	
May 1958	4.0, 4.0, yes	3.5, 2.0, yes 4.0, 4.0, yes	4.0, 4.0, yes
June 1958		3.0, 0.0, yes–	3.5, 0.0, yes–
July 1958		4.0, 4.0, yes	3.0, 0.0, slight– 4.0, 3.5, slight
August 1958			0.5, 0.0, slight– 1.5, 0.0, slight
September 1958			0.0, 0.0, no
October 1958			3.0, 0.0, slight– 3.5, 2.0, yes
November 1958	2.5, 0.0, no		3.5, 3.0, yes
December 1958	3.0, 1.0, slight		4.0, 3.0, yes
January 1959	4.0, 3.0, yes		2.0, 0.0, slight–
February 1959	2.0, 0.0, slight– 4.0, 4.0, yes		

* The first number refers to stage of lateral cement glands, the second to stage of midventral glands, "slight" refers to condition of oviducal plug.

– carrying eggs or young

ing season (group C) may mature and become ovigerous at a small size in late June, July and August. In 1958 group C could be subdivided into two parts: (1) those animals hatched near the beginning of the breeding season, and (2) those hatched near the end of it. Due to drought conditions, the 1959 group C corresponded only to (2) of the 1958 group. The larger of the group C animals may have a second brood in October. A very few of the smaller group C animals may survive the uary. The June young (Group A) do not winter and produce a second brood in Janattain sufficient size to reproduce until the following January and February. They reproduce only once. The young produced in late July, August and October (group B) reach maturity and may produce young in March and reproduce again in June.

The peaks of reproductive activity thus produced are in fair agreement with those published by Penn (1950) for *C. shufeldti.* His data included animals collected throughout the state of Louisiana. As local conditions vary, the expected flattening of the curve occurs; the peaks he reports are not quite so high nor the dips so sharp.

This is the shortest life cycle reported for any crayfish; others studied have been those of larger animals that required a longer time to reach maturity and lived longer in an adult condition (Hobbs, 1942; Penn, 1943; Smith, 1953). In these studies length was the only basis for separating different age classes. In *C. shufeldti* at least, length alone is not sufficient, as two distinct age groups, clearly demarcated by weight, may have overlapping cephalothorax length measurements. Thus weight and length together must be used to define age groups accurately in this small shortlived species.

Influence of Environmental Factors

Light.—To determine the effect of day-length on the reproductive cycle, five groups of crayfish were maintained under different photoperiods from June 10 through September 15, 1958. The animals were collected on June 1, 4, 6, and 9, and not separated as to collection date. Males and females were present in each group. The crayfish were contained in covered rectangular stainless steel tanks, 49 x 37 cm, kept side by side in an air-conditioned laboratory. Water was changed weekly; the animals were not fed during the course of the experiment. Illumination was provided in each tank by one frosted 10-watt bulb suspended 20 cm above the water surface. Intensity of illumination at the surface of the water was approximately 40-45 foot-candles. Duration of light was controlled by automatic time clocks set to provide respectively 0, 6, 12, 18, and 24 hours of illumination daily. Illumination began at 6 AM.

Beginning July 1 and every fifteen days until September 15 a random sample of animals was removed from each tank and preserved in Bouin's fixative for future examination. As both group B and group C individuals were present during the early part of the experiment, the animals were weighed and cephalothorax length measured to separate the groups. The ovaries were dissected out, weighed and largest oocyte diameters measured. The results are summarized by group in Tables 5 and 6. Stage of cement gland development is also recorded in these tables.

In group B animals, darkness caused an initial increase in ovarian size and this increment was maintained through August 15, the last date on which animals of this group were present in the population. Exposure to six-hour day-length also caused increase in ovarian size but this was followed by a decrease in weight. For 12-, 18-, and 24-hour day-lengths there was a decrease in ovarian weight. However, with 18 and 24 hours illumination daily this initial decrease was followed by a weight increase.

The same general sequence was shown by animals of group C (Fig. 4). At the first sampling on July 1 these animals had just molted to a mature size and there was an increase in ovarian size in 15 days under all lighting conditions (Fig. 4, B, H, N, T, Z). A change from the situation in group B was that animals receiving a 12-hour photoperiod showed as great an increase as caused by complete darkness. By August 1 the ovarian weight of the animals in darkness was stabilized, while that of those under six-hour illumination showed an additional increase, and the other groups a decrease. By the middle of August the animals receiving 18 to 24 hours of light daily (Fig. 4, V, BB) showed an increase in

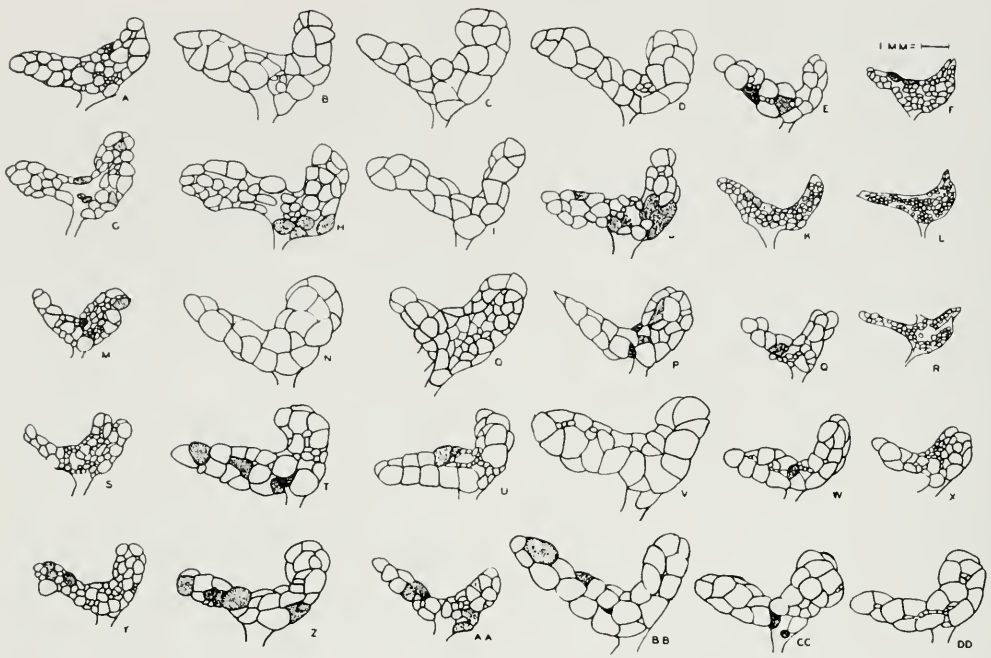

Figure 4. Changes in ovarian condition of group C crayfish in response to different photoperiods. 0 hours illumination, A-F; 6 hours, G-L; 12 hours, M-R; 18 hours, S-X; 24 hours, Y-DD.

ovarian weight while the others exhibited a decrease. In both September samples groups under all illuminations showed a decrease in ovarian weight, the most rapid decrease occurring in animals in darkness and the least rapid in those under continual illumination.

The cement glands generally followed the pattern shown by the ovaries (Tables 5 and 6). However, the correlation was not per-

TABLE 5.
Changes in ovarian condition due to different photoperiods: group B animals.

Day-Length (Hrs.)	Date	Ovarian weight range	Ovarian weight mean	Oocyte diameter range	Oocyte diameter mean	Cement gland stage	No. of animals
0	7-1	11.5-13.9	12.5	1.50-1.75	1.68	2.5, 0.0, slight	9
	7-15	13.1-15.6	14.0	1.50-1.81	1.70	2.5, 0.0, yes	8
	8-1	13.0-14.9	13.8	1.50-1.70	1.60	3.5, 1.5, yes	5
	8-15	13.2-14.7	13.9	1.48-1.55	1.51	3.0, 1.0, yes	5
6	7-1	10.5-12.5	11.7	1.52-1.67	1.60	2.5, 0.0, yes	8
	7-15	10.9-13.0	12.2	1.50-1.73	1.60	3.0, 2.0, yes	6
	8-1	7.7- 8.2	8.0	1.28-1.36	1.32	2.0, 0.0, yes	4
12	7-1	10.5-11.9	11.2	1.51-1.70	1.62	2.5, 0.0, yes	7
	7-15	9.7-10.8	10.3	1.41-1.54	1.47	2.5, 0.0, yes	8
	8-1	7.9- 9.8	8.1	1.15-1.26	1.20	3.0, 1.0, yes	6
18	7-1	10.2-12.0	11.1	1.56-1.65	1.60	2.5, 0.0, yes	9
	7-15	9.1-10.9	9.7	1.20-1.47	1.38	3.5, 2.0, yes	7
	8-1	9.2-11.1	10.5	1.40-1.49	1.44	3.5, 2.0, yes	8
	8-15	14.4-16.9	15.9	1.44-1.62	1.55	4.0, 1.0, slight	5
24	7-1	9.2-10.8	10.1	1.50-1.59	1.55	2.5, 0.0, slight	5
	7-15	6.9- 7.9	7.6	1.19-1.25	1.22	2.5, 0.0, yes	6
	8-1	8.5-10.7	9.5	1.30-1.39	1.35	3.0, 1.0, yes	7

TABLE 6.
Changes in ovarian condition due to different photoperiods: group C animals.

Day-Length (Hrs.)	Date	Ovarian weight range	Ovarian weight mean	Oocyte diameter range	Oocyte diameter mean	Cement gland stage	No. of animals
0	7-1	2-1-3.8	3.2	0.85-0.91	0.91	2.5, 0.0, slight	6
	7-15	6.6-7.8	7.2	1.13-1.40	1.30	2.5, 0.0, yes	7
	8-1	6.0-7.9	6.9	1.26-1.40	1.33	3.5, 2.0, yes	8
	8-15	6.0-7.8	6.7	1.26-1.43	1.34	3.0, 2.0, yes	6
		1.9-2.1	2.0*	0.65-0.69	0.67		2
	9-1	1.4-3.0	2.1	0.63-0.91	0.78	1.0, 0.0, no	10
	9-15	0.7-1.2	0.9	0.35-0.43	0.38	0.0, 0.0, no	10
6	7-1	2.0-3.4	2.8	0.71-0.93	0.80	2.5, 0.0, slight	7
	7-15	4.6-6.0	5.3	0.92-1.12	1.00	2.0, 0.0, yes	9
	8-1	6.4-7.8	7.1	1.21-1.42	1.31	3.0, 2.0, yes	8
	8-15	3.9-5.7	4.9	0.67-0.91	0.81	1.5, 0.0, slight	7
	9-1	1.0-2.2	1.6	0.35-0.53	0.43	0.5, 0.0, no	6
	9-15	0.5-1.0	0.8	0.25-0.33	0.30	0.0, 0.0, no	5
12	7-1	2.0-3.0	2.4	0.60-0.71	0.66	2.0, 0.0, slight	8
	7-15	6.0-8.4	7.4	1.31-1.56	1.40	3.5, 3.0, yes	7
	8-1	5.0-6.1	5.6	0.89-1.06	0.97	3.5, 3.0, yes	8
	8-15	2.8-3.5	3.2	0.81-0.97	0.89	3.5, 2.0, no	9
	9-1	1.5-2.3	1.9	0.68-0.76	0.73	0.5, 0.0, no	6
	9-15	0.3-0.9	0.5	0.21-0.30	0.25	0.0, 0.0, no	10
18	7-1	1.9-3.3	2.4	0.42-0.75	0.60	2.5, 0.0, slight	6
	7-15	5.2-6.4	5.9	0.98-1.08	1.04	3.5, 1.0, slight	7
	8-1	3.4-4.5	3.7	0.87-0.98	0.93	3.0, 0.0, no	7
	8-15	6.5-8.0	7.4	1.32-1.47	1.40	4.0, 4.0, yes	6
	9-1	2.5-4.2	3.2	0.78-0.91	0.85	3.0, 1.0, slight	8
	9-15	1.0-3.0	1.7	0.69-0.75	0.72	3.0, 1.5, slight	11
24	7-1	1.5-3.0	2.1	0.69-0.76	0.73	2.5, 0.0, slight	9
	7-15	3.9-5.4	4.5	0.85-0.95	0.90	3.0, 0.0, yes	8
	8-1	2.5-3.9	3.2	0.99-1.10	1.06	2.0, 0.0, yes	8
	8-15	6.3-7.7	7.0	1.41-1.53	1.47	4.0, 4.0, yes	8
		2.0-2.4	2.2*	0.85 -	0.85		2
	9-1	3.2-5.0	4.1	1.06-1.17	1.12	3.0, 2.0, yes	9
	9-15	1.1-3.0	2.0	0.72-0.84	0.78	3.0, 2.0, slight	10

* ovigerous

fect. In darkness there was a time lag in the maximum gland development and then the glands decreased while the ovarian weight remained steady. In group B animals from July 1 to July 15 there was no decrease in cement gland stage although three of the groups showed decreased ovarian weight and oocyte diameter.

The longer the photoperiod, the greater the cement gland development; only under 18- and 24-hour day-lengths did the glands reach the fully mature 4.0, 4.0 condition. For approximately the same ovarian weight the glands of animals exposed to long day-lengths were more fully developed.

The only animals to lay eggs during the course of the experiment were animals maintained in constant darkness and in continual light. Two animals of each of these groups were ovigerous on August 15 (Table 5). While this fact is interesting the samples were not large enough to determine the significance of egg-laying by members of these two groups.

Duration of light also had an effect on the ovaries of immature animals of 7.0, 7.5, and 8.0 mm cephalothorax length (Table 7). These animals were present only in three tanks; 0, 12, and 18 hours light. Although these data are meager, the average representing never more than five animals, the greater ovarian maturation of those exposed to 18 hours illumination was very marked. There was a steady increase in ovarian maturity as exposure to long day-length continued and by September the

TABLE 7.
Average ovarian weights of immature animals exposed to 0, 12, and 18 hours light daily from June 10 to September 15, 1958.

Date	0 hours Ovarian weight	0 hours Oocyte diameter	12 hours Ovarian weight	12 hours Oocyte diameter	18 hours Ovarian weight	18 hours Oocyte diameter
7-1	.5	.20	.5	.20	.5	.20
7-15	.4	.25**	.7	.25*	.6	.30
8-1	.5	.25	.5	.20**	.9	.43*
8-15	.5	.21*	.4	.19	1.2	.69
9-1	.4	.20**	.3	.13	2.0	.95
9-15	.4	.20	.3	.14**	2.4	1.06

* two animals only ** three animals only
(The other measurements represent four or five animals)

oocytes had a diameter similar to that found in larger animals containing ovaries of 3 to 6 mg. A few crayfish of 6 mm cephalothorax length were present in the tanks, but ovaries of animals this small were indifferent to light conditions. They remained tiny, transparent threads containing minute oocytes.

Cement gland development paralleled ovarian development. The glands of animals in darkness and under 12 hours illumination remained undeveloped. Those under 18 hours illumination developed gradually to stage 3.0 (lateral glands), 1.0 (midventral glands) at the termination of the experiment. This degree of development was closely comparable to the stage 3.0, 1.5 of adult animals under the same conditions. This is in contrast to the results of G. C. Stephens (1952) who reported no increase in cement gland development in juvenile *O. virilis* subjected to long photoperiods. As the ovaries also remained in a juvenile condition, the animals used by Stephens might be comparable to the 6 mm *C. shufeldti* which were too young to respond.

This effect of photoperiod may explain why the ovaries of animals measuring 8.5 mm cephalothorax length were found in a mature condition only in July and August; exposure to long day-length for a period of time may be necessary to bring them to maturity. However, natural day-length is not long enough to cause animals smaller than 8.5 mm to mature.

A similar series of photoperiod experiments were undertaken beginning September 30, 1959. At the beginning of the experiment all crayfish possessed undeveloped ovaries. The only crayfish to survive an unscheduled spraying with insecticide were nine of the animals in darkness. Four of these were sacrificed immediately. October 28, and the remainder on November 28. These results are included because the response of the crayfish to darkness differed from the response in the summer experiments. The ovaries did not develop (Table 8. The small weight increase in November was due to proliferation of new oocytes and not to maturation.

Temperature.—To determine the effect of

TABLE 8.
Changes in ovarian condition due to darkness: placed in darkness September 30, 1959.

Date	Ovarian weight range	Ovarian weight mean	Oocyte diameter range	Oocyte diameter mean
Darkness				
10-28	0.9- 2.9	2.0	0.35-0.62	0.47
11-28	1.8- 5.3	3.4	0.33-0.65	0.48
Control (from Table 3)				
9-15	0.7- 1.6	1.1	0.37-0.61	0.48
10-15	8.0-10.9	9.3	1.15-1.23	1.18
11-15	12.8-15.8	14.3	1.20-1.38	1.28

TABLE 9.
Changes in ovarian condition due to different temperatures: placed at constant temperature November 5, 1958.

Date	Ovarian weight range	Ovarian weight mean	Oocyte diameter range	Oocyte diameter mean	Cement gland development
5.5-7.5°C.					
11-22	1.4-2.8	2.1	0.60-0.74	0.67	1.0, 0.0, no
11-29	1.9-4.5	2.8	0.60-0.95	0.81	1.0, 1.0, no
12-6	2.5-6.1	4.0	0.67-1.00	0.82	3.0, 2.0, yes
12-20	6.3-8.5	7.4	1.06-1.28	1.20	3.0, 2.0, yes
14.5-19.5°C.					
11-22	4.0-5.5	4.8	0.78-0.95	0.87	2.0, 0.5, no
11-29	4.5-7.5	6.0	0.87-1.13	1.00	2.5, 1.0, yes
12-6	5.0-8.2	6.4	0.92-1.13	1.00	2.5, 2.0, yes
12-20	6.5-9.5	7.6	1.23-1.33	1.27	3.0, 2.0, yes
29.5-30.5°C.					
11-22	4.6-6.7	5.6	1.24-1.30	1.27	2.5, 1.0, yes
11-29	1.4-5.5	3.3	0.58-1.07	0.81	2.0, 0.5, slight
12-6	0.8-2.1	1.5	0.43-0.83	0.65	1.0, 0.0, slight
12-20	0.3-1.2	0.8	0.17-0.53	0.33	1.0, 0.0, no

temperature on the reproductive cycle of *C. shufeldti*, groups of crayfish were maintained at 6.5°C, 17°C, and 30°C for a period of one and one-half months. The same type tank and lighting arrangement as used in the light experiments was employed. All groups received 12 hours light per day beginning at 6 AM. The 30°C temperature was maintained by a hot water bath equipped with thermostat, the 17°C by a refrigerator, and the 6.5°C by a constant temperature room. Variation in temperature was recorded by maximum-minimum thermometers. The mortality of the 6.5°C group was initially high and then decreased to a low level. Very few died in the other two groups. Both males and females of groups B and C were present in each tank but only group C females were dissected. The animals were not fed during the course of the experiment.

The ovaries were dissected in van Harreveld's saline solution, which is isotonic to the blood of fresh water crustaceans (van Harreveld, 1936). The ovaries were drawn in dorsal view. The wet weight of each ovary was recorded. However, as a constant amount of saline was not transferred with each ovary, they were later reweighed after preservation so these weights would be comparable to those of other experiments.

Table 9 summarizes the sequence of ovarian changes under the three different temperatures and Figure 5 pictures ovarian condition. The animals were placed at constant temperature on November 5 and sampled at frequent intervals. At 17 ± 2.5°C the ovarian development was similar to that of animals collected in the field. The ovaries from animals in the December 15, 1958, field collection averaged 7.8 mg and had an average oocyte diameter of 1.30 mm (Table 3) while the experimentals dissected December 20 averaged 7.6 mg with an oocyte diameter of 1.27 mm.

The cooler temperature of 6.5 ± 1°C initially slowed development. However, the rate of development gradually increased so that by the end of the experiment the inhibition due to cold had been overcome and the condition of these ovaries and that of the 17°C group was similar.

Placing animals at an elevated temperature, 30 ± 0.5°C, caused a rapid increment in the ovarian size followed by deterioration with resorption of already formed oocytes and only limited proliferation of new ones.

At reduced temperatures there was a continual proliferation of new oocytes (Fig. 5, A-D) scattered among the maturing ones. At 17°C this proliferation was not so apparent (Fig. 5, E-H) and at 30°C the development of new oocytes was greatly slowed, the ovary being filled largely with a transparent interstitial material (Fig. 5, I-L).

The series of experiments with temperature was repeated in February-April, 1959.

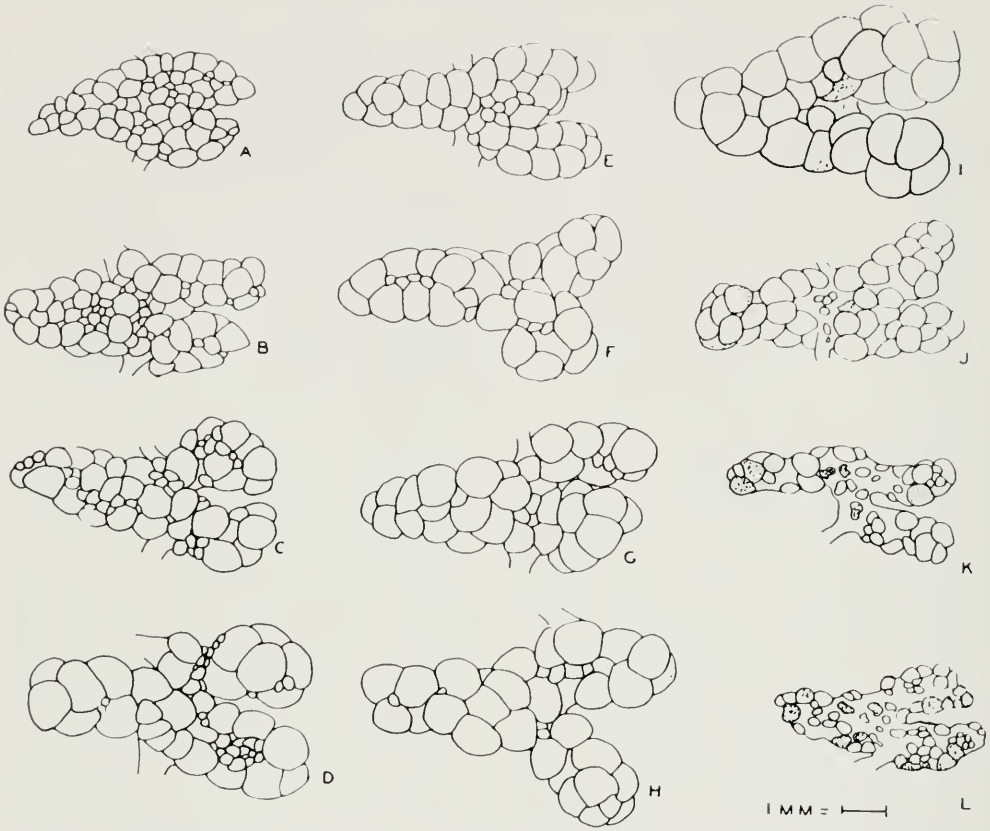

Figure 5. Sequence of changes in ovarian development under various temperatures. 7°C, A-D; 17°C, E-H; 30°C, I-L.

The low temperature was raised to 8.75 ± 1.25°C to lower the mortality that occurred at 6.5°. The high temperature was 28.75 ± 0.25°C rather than 30 ± 0.5°C. Only group A animals that were ovigerous at the start of the experiment were used. To minimize the possibility of the animals at higher temperatures resorbing the ovary as a food source because of their higher metabolic rate, strands of *Elodea* were placed in the tanks and some canned cat food added every other day.

Even with what was assumed to be an abundant food supply, the experimental results (Table 10) were essentially similar to those previously reported; a slowing of development at low temperatures and a sudden increase followed by resorption at high temperatures. However, the lag in ovarian development at low temperature increased with time rather than decreased. Also, the regression at a higher temperature proceeded at a slower rate and interstitial material did not replace the oocytes to as great an extent.

In the winter experiment cement gland development closely paralleled ovarian development, increasing or decreasing with ovarian development (Table 9). In the spring, however, cold temperature did not inhibit gland development; the glands of animals exposed to temperatures of 9°C were as fully developed as those of animals at 17°C (Table 10). The glands of animals at 29°C remained almost stationary at stage 3.0, 0.0.

Temperature affected the development of attached eggs and developing young. All animals carried eggs in which little development was apparent at the start of the 1959 experiment. At 29°C only two animals carried young at the time of the first sampling (18 days after the experiments began). The rest exhibited only empty egg cases because the young had already left the

TABLE 10.
Changes in ovarian condition due to different temperatures: placed at constant temperature February 28, 1959.

Date	Ovarian weight range	Ovarian weight mean	Oocyte diameter range	Oocyte diameter mean	Cement gland stage
28.5-29.0° C.					
3-17	2.4- 2.9	2.7	0.55-0.74	0.65	3.5, 0.0, yes
3-25	2.5- 4.9	3.9	0.67-0.80	0.73	4.0, 2.0, yes
4-1	3.7- 5.6	4.8	0.87-0.97	0.93	4.0, 3.0, yes
4-10	4.8- 5.6	5.2	0.87-0.98	0.94	4.0, 2.0, yes
16-19° C.					
3-17	1.7- 3.4	2.5	0.48-0.67	0.53	2.5, 0.0, slight
3-25	4.1- 5.6	4.9	0.76-0.87	0.81	3.5, 1.0, yes
4-1	6.5- 7.5	7.1	0.90-1.06	0.97	3.5, 1.0, yes
4-10	11.9-13.7	12.8	1.06-1.23	1.12	4.0, 2.0, yes
7.5-10.0° C.					
3-17	3.8- 7.4	5.4	0.87-1.20	1.08	3.0, 0.0, yes
3-25	3.0- 6.8	4.7	0.83-1.20	0.97	3.0, 0.0, yes
4-1	3.0- 5.0	4.0	0.67-0.95	0.80	3.0, 0.0, yes
4-10	2.8- 3.6	3.2	0.71-0.85	0.78	2.5, 0.0, slight

pleopods. At 17°C, development was slower and a few young were still present on the pleopods after 22 days. A temperature of 9°C greatly slowed development; eggs were still present at the termination of the experiment, a total of 42 days. A few of these animals were then returned to room temperature and many of the eggs hatched in a few days.

Discussion. — Temperature, duration of

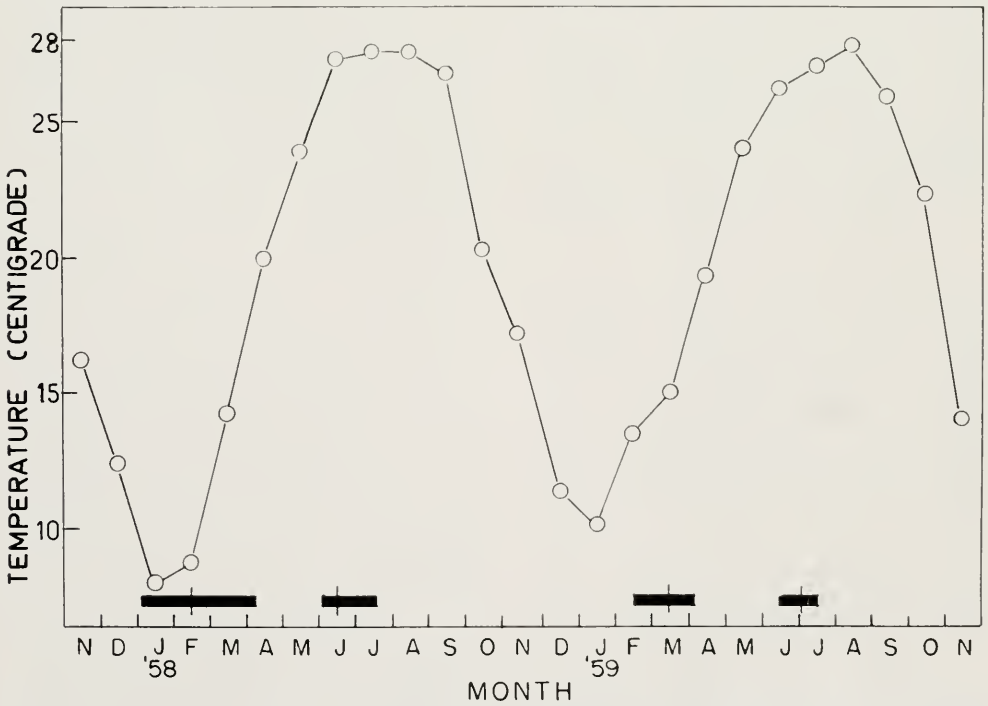

Figure 6. Annual variation in temperature at Slidell, La., expressed in terms of mean monthly temperature. The solid bars at the base of the figure indicate the period during which 60 percent or more of the females were brooding. The vertical line through each bar indicates the peak reproductive period.

light, and rainfall influence the reproductive cycle of *Cambarellus shufeldti.* The peaks of reproductive activity occur (1) in the coldest portion of the year, slightly after the shortest day-length, and (2) at the beginning of the hottest season which is also the time of longest day-length. Figure 6 shows the variation in mean monthly temperature throughout the year in the vicinity of the collection site. As the U. S. Weather Bureau does not maintain a temperature station at Pearl River, the temperatures recorded are from Slidell, Louisiana, eight miles southwest of the collection area. Figure 7 gives the variation in day-length throughout the year at New Orleans, Louisiana, 26 miles southwest of the collection site and at sea level rather than 29 feet elevation. The times during which 60 percent or more of the adult female population carried eggs or young are indicated by the horizontal bars on the figures.

The decreasing temperatures and shortening day-lengths of late fall and early winter stimulate proliferation of new oocytes and inhibit ovarian maturation. Short days tend to stabilize the oocytes as they develop rather than allow the quicker cycling of yolk deposition and resorption induced by long photoperiods. Gradually the inhibition induced by cold weather and short day length is lost, the ovaries mature and egg laying occurs.

As temperature is rapidly increasing toward the summer plateau, the young of the year grow and mature quickly. The ovaries of adult animals develop rapidly and there is a sharp peak of reproductive activity in June. Long day-lengths accelerate the cyclic activity of the ovary.

While the ovary in June and July grows very quickly after expelling eggs, continued high temperature induces yolk resorption and the ovaries regress until September, when they approximate the condition seen in immature animals. Day-length has been increasing from March through June and smaller animals are finally stimulated to mature and lay eggs in July and August before their ovaries also regress. The strong inherent tendency for the late summer decline is shown by the series of experiments with day-length.. The ovary regressed under all illuminations in September. That this may be associated with a general metabolic factor is shown by the fact that animals collected from the field in August and September exhibited less in body weight increase than in any other months during the year.

As cooler weather and shorter photoperiods set in, the ovaries begin to grow again, new oocytes are proliferated and a few animals are able to lay eggs before colder weather slows maturation of oocytes. The June young do not reach maturity before cold weather begins and are not able to reproduce until the following year.

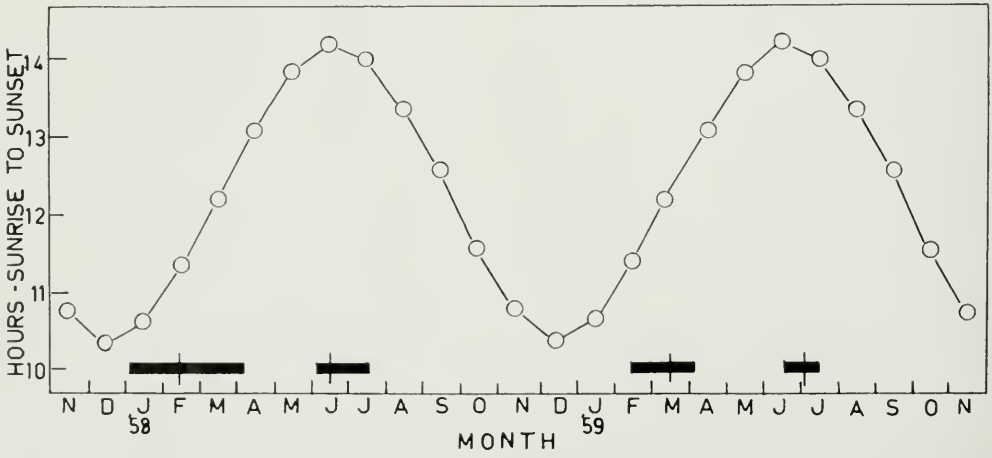

Figure 7. Annual variation in photoperiod at New Orleans, La., expressed as monthly mean number of hours from sunrise to sunset. The solid bars at the base of the figure indicate the period during which 60 percent or more of the females were brooding. The vertical line through each bar indicates the peak reproductive period.

Rainfall can influence the reproductive cycle to the extent that animals may be forced to burrow if rainfall is scanty. This occurred in December 1958 - January, 1959. The forced burrowing of the crayfish placed them in complete darkness and may give an extra spurt to ovarian development. The combination of cold and darkness, plus forced retention of the oocytes may account for the large size reached by the ovaries before the animals laid eggs in 1959.

Temperature has another effect on the reproductive cycle. Reproductive activity throughout the year was determined by the percentage of females carrying eggs or young. The time required for the eggs to hatch and for the young to leave the pleopods is inversely related to temperature. Mean monthly temperatures may vary from year to year and were lower during the 1958 winter breeding season than in 1959 so that, in effect, the period of peak reproductive activity was extended in 1958.

Lack of rainfall augmented the temperature effect in 1958-1959 by forcing the animals to burrow and thus delayed their breeding season. By the time standing water was again present, temperatures were warmer and the time required for the young to develop shorter; thus shortening further the span of the reproductive peak in 1959.

The data presented here agree with that of G. J. Stephens (1952) and Suko (1958). Stephen's experiments revealed that darkness stimulated maturation of oocytes and may inhibit cyclic activity, while increased light accelerated the cycle. Suko reported that oocytes of *Procambarus clarkii* kept in darkness for two to three months after oviposition displayed no histological change in spite of the elongation of their ovaries (indicating oocyte proliferation without maturation). Ovaries of animals kept further over this period developed earlier than those of controls. Animals kept in darkness immediately before breeding and egg-laying showed degenerative changes in the ovary.

C. shufeldti in the present experiments also showed varied response to lack of light depending on the time of year.

Further experiments on the effects of light are needed. Also in the present experiments, there was little difference between ovaries of animals exposed to 18 or 24 hours of light. The greatest day-length in New Orleans is 14 hours. How much the photoperiod can be reduced before distinct differences appear would be interesting to determine.

Summary

1. The reproductive cycle of the female dwarf crayfish, *Cambarellus shufeldti,* is defined. While females carrying eggs on their pleopods can be found almost any time throughout the year, there are two peaks of reproductive activity, the largest in late winter-early spring, the other in June.

2. Three distinct groups of adult females appear in the population during the year. Individual crayfish probably live no longer than a year and no longer than six months as adults. During this time an animal may reproduce twice.

3. Increase of photoperiod induces a more rapid cycling of the maturation and resorption of oocytes. Decrease of day-length tends to stabilize the ovary in a mature condition at one time of year, while at another time tends to allow increased proliferation of oocytes without maturation.

4. Lowered temperature tends to slow maturation of oocytes while elevated temperatures cause a quick maturation followed by disintegration of the ovary.

5. *C. shufeldti* appears to have an inherent rhythm of reproductive activity, as shown by varied response to experimental lighting and temperature conditions at different times during the year.

References Cited

Fingerman, Milton 1957. Regulation of the distal retinal pigment of the dwarf crayfish, *Cambarellus shufeldti. Jour. Cell. and Comp. Physiol.,* 50: 357-370.

---------- and Mildred E. Lowe 1957. Hormones controlling the chromatophores of the dwarf crayfish, *Cambarellus shufeldti:* their secretion, stability and separation by filter paper electrophoresis. *Tulane Stud. Zool.,* 15(7): 151-171.

Giese, Arthur C. 1959. Annual reproductive cycles of marine invertebrates. *Ann. Rev. Physiol.,* 21: 547-576.

Hobbs, Horton H. Jr. 1942. The crayfishes of Florida. *Univ. Fla. Publ., Biol. Sci. Ser.,* 3: 1-179.

Korringa, P. 1947. Relations between the moon and periodicity in the breeding of marine animals. *Ecol. Monogr.,* 17: 347-381.

---------- 1952. Recent advances in

oyster biology. *Quart. Rev. Biol.*, 27: 266-308, 339-365.

Lees, A. D. 1955. *The Physiology of Diapause in Arthopods.* Cambridge Univ. Press, 150 pp.

Loosanoff, V. L. and H. C. Davis 1952. Repeated semi-annual spawning of northern oysters. *Science*, 115: 675-76.

Penn, George Henry 1942. Observations on the biology of the dwarf crawfish, *Cambarellus shufeldti* (Faxon). *Amer. Midl. Nat.*, 28: 644-647.

1943. A study of the life history of the Louisiana red crawfish, *Cambarus clarkii* Girard. *Ecology*, 24: 1-18.

1950. The genus *Cambarellus* in Louisiana. *Amer. Midl. Nat.*, 44: 421-426.

Smith, Elsie Wayne 1953. The life history of the crawfish *Orconectes (Faxonella) clypeatus* (Hay). *Tulane Stud. Zool.*, 1: 77-96.

Stephens, Grover C. 1952. The control of cement gland development in the crayfish *Cambarus*. *Biol. Bull.*, 103: 242-258.

Stephens, Gwen J. 1952. Mechanisms regulating the reproductive cycle in the crayfish *Cambarus*. I. The female cycle. *Physiol. Zool.*, 25: 70-83.

Suko, Tetsuya 1958. Studies on the development of the crayfish. V. The histological changes of the developmental ovaries influenced by the condition of darkness. *Sci. Rept. Saitama Univ.*, B-3(1): 67-78.

Van Harreveld, A. 1936. A physiological solution for freshwater crustaceans. *Proc. Soc. Exper. Biol. Med.*, 34: 428-432.

Abstract

The reproductive cycle of the female crayfish *Cambarellus shufeldti* was defined. Field collections were made at least once a month from November 1957 through November 1959. While females carrying eggs on their pleopods could be found almost any time throughout the year, there were two peaks of reproductive activity, the largest in late winter-early spring, the second in June-July. These periods occurred shortly after the coldest portion of the year and shortest day-length and at the beginning of the hottest portion which is also the time of longest day-length.

Three distinct groups of adult females appeared in the population during the year as determined by total body weight and cephalothorax length. Individual crayfish probably lived no longer than a year and no longer than six months as adults. During this time an animal could reproduce twice.

Under increased photoperiod a more rapid cycling of maturation and resorption of oocytes occurred. Decreased day-length tended to stabilize the ovary in a mature condition or to allow increased proliferation of oocytes without maturation, depending on time of year.

Lowered temperature tended to slow maturation of oocytes while at elevated temperatures there was quick maturation followed by disintegration of the ovary.