OBSERVATIONS ON THE LIFE HISTORIES AND FEEDING OF *CINYGMA INTEGRUM* EATON AND *IRONODES NITIDUS* (EATON) (EPHEMEROPTERA: HEPTAGENIIDAE)[1]

C. R. D. Pereira[2] and N. H. Anderson

Entomology Department,
Oregon State University
Corvallis, OR 97331

[1]Technical Paper No. 6421, Oregon Agriculture Experiment Station.

[2]Present Address: Entomology Department, Purdue University, West Lafayette, IN 47906.

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ABSTRACT

Larval growth and development of *Cinygma integrum* and *Ironodes nitidus* were studied for one year at Berry Creek, Benton Co., Oregon. *C. integrum* has a univoltine, slow seasonal type of life cycle. Two cohorts, having different rates of maturation at any one period of time, were observed. The larger cohort began development in the summer while the smaller cohort began development in the fall. This phenomenon of two cohorts in the life cycle appeared to be related to temperature regime and stability of the environment at the study site. *Ironodes nitidus* has a univoltine, fast seasonal type of life cycle with only the egg stage being present in the summer. The proportion of *C. integrum* larvae on wood substrates in riffle areas was 76% whilst that of *I. nitidus* was 43%. Both species were found to ingest large quantities of fungal material especially during the winter.

INTRODUCTION

Amongst the mayflies associated with wood debris in western streams we have identified the heptageneiids, *Cinygma integrum* Eaton and *Ironodes nitidus* (Eaton) as the principal species that ingest wood particles (Dudley and Anderson, 1982; Pereira *et al.*, 1982). In the Dudley and Anderson study, *I. nitidus* was included as part of the genus *Epeorus*. This paper provides data on the life histories of the two species and quantification of substrate preferences and feeding habits of the larvae from a study at Berry Creek, near Corvallis, Oregon (Pereira, 1980).

The genus *Cinygma* occurs in the Holarctic and Oriental regions and three of the four described species occur in the Pacific Northwest (Lehmkuhl, 1979). *C. integrum* is the only species in which the larva is described. Life-history data are limited to emergence records (Eaton, 1885; McDunnough, 1933; Allen, 1955) and seasonal occurrence of larval size classes (Lehmkuhl, 1979). *I. nitidus* is distributed throughout the Pacific Northwest. The life cycle of this species was studied by Lehmkuhl (1968) at Oak Creek, Benton Co., Oregon.

MATERIALS AND METHODS

Field studies were conducted from July 1979 to July 1980 at Berry Creek, Benton Co., Oregon. This second-order stream drains the northern slope of MacDonald Forest in the eastern foothills of the Coast Range. Sampling was carried out on the South Fork of Berry Creek in the area brought under controlled flow (Warren *et al.*, 1964). The stream temperatures do not show dramatic fluctuations because the study area is almost completely covered by a deciduous canopy. Because of the flow control, the stream does not experience the normal winter freshets. This situation has existed for 20 years, consequently the channel contains an excessive amount of fine organic material and an abundance of large and small wood debris.

Monthly samples were taken for all mayflies for one year from wood debris, rocks, and "composite substrates" in riffle areas. Wood debris (>6 cm dia) and stones (>10 cm dia) were picked from the stream, washed thoroughly into a bucket, and the contents filtered through a sieve of mesh size 8 meshes/mm. "Composite substrate" samples (henceforth referred to as kick samples) were collected by disturbing the bottom substrate and collecting the dislodged material with an aquatic net (4 meshes/mm) which was held downstream of the disturbed area. Approximately $1m^2$ of each substrate type was sampled each month.

To determine the life cycle pattern, the larvae were measured for both body length and head-capsule width, and placed in size classes. However, as size is partially dependent on environmental conditions, the larvae were also partitioned into physiological age classes or developmental stages similar to those used by Lehmkuhl (1969). For size classes, body length (excluding cerci) was measured to the nearest 0.1mm. Each size class was 1mm in range, except that all larvae <2mm were grouped together. The developmental stages were defined as: Stage A, no wing pads developed; Stage B, fore-wing pads present but no hind-wing pads; Stage C, both fore- and hind-wing pads present but the former do not completely cover the latter; Stage D, fore-wing pads completely cover the hind-wing pads; Stage E, dark wing pads present.

Adults were collected in conical emergence traps enclosing an area of $0.25m^2$. In 1979 the mayflies were obtained as part of a larger study which included traps set over both riffles and pools from April to August. In 1980, two traps were placed on riffles over each of wood, stone, and "composite substrates" from February to June. Adults from the emergence traps were used to determine sex ratio, longevity and fecundity.

FEEDING HABITS

This part of the study was directed mostly towards studying the impact of feeding on wood decomposition and mineralization processes. Larvae from monthly wood substrate samples (preserved in 70% alcohol) were dissected and the contents of the foregut were examined under a microscope at 450 X magnification. Three different fields were observed for each specimen examined. The following food categories were selected: (1) detritus—which consisted of wood, leaf material and other unidentifiable material; (2) fungal elements—spores and mycelia; (3) diatoms and filamentous algae, and (4) mineral particles. Detritus was subdivided into wood and leaf material but this was inadequate for quantitative distinction since it was sometimes difficult to decide if the material was wood or of non-woody origin. Wood was distinguished by the presence of tracheids, highly lignified cell walls, pits and fibers. Spores were recognized by the thick cell wall and shape, and the mycelia by the color and consistency of the cell wall and clamp connections.

RESULTS AND DISCUSSION

ADULTS

About 40 adults each of *C. integrum* and *I. nitidus* were collected from emergence traps. Both species had extended emergence periods: *I. nitidus*, February-August; *C. integrum*, March-August. Though the numbers are too low for definitive comparisons, it seemed that *C. integrum* emerged over a range of velocities from pools to riffles whereas *I. nitidus* only emerged from riffles. Both species used wood substrates as emergence platforms, with about 70% of the *I. nitidus* and 45% of the *C. integrum* adults being collected from traps set over this substrate. The sex ratio of *I. nitidus* was 1:1, whereas that of *C. integrum* was close to 2♀♀:1♂, based on collections of adults and mature larvae.

Adults of *I. nitidus* were observed swarming at about 2:30 pm. The swarm contained 25-35 individuals flying at 3-5 m above the water. No swarms of *C. integrum* were seen nor were any adults collected by sweeping the vegetation. It is likely that the subimagos rest high in the trees as those that emerged in the laboratory immediately flew to the ceiling. Two subimagos of *C. integrum* emerged on the field around 3 pm, while in the laboratory emergence most often occurred in late afternoon.

Adults of *C. integrum* lived for 2-10 days in the laboratory; those with the longest life-span had been held at 10°C compared with room temperature for short-lived ones. *I. nitidus* adults lived for 3-5 days at 10°C.

Average fecundity of field-collected females was 1148 for *C. integrum* (N=6) and 1611 for *I. nitidus (N*=2).

LIFE CYCLE PATTERNS

CINYGMA INTEGRUM. The life cycle of this species is somewhat difficult to interpret because there is no clearly defined cohort showing progressive growth. Larvae in the <2 mm size class were present in several months (Fig. 1), although they were most common in June-July and November-December. Eggs apparently hatch over an extended period from summer to late fall resulting in continuous recruitment of the larvae, expressed as <2 mm size class (Fig. 1) and development stage A (Fig. 2). The long emergence period of adults (Fig. 2) indicates that oviposition will be spread over much of the spring and summer.

C. integrum is a cool-adapted species. Small larvae grow slowly during the summer and fall and the major growth period occurs during the winter and spring. The developmental stage analysis (Fig. 2) suggests there is a second smaller cohort of stage A and B larvae in January-February, but that these converge with the main cohort in the spring. By using modal values of head capsule width, (Fig. 3) it appears that the late-emerging larvae in November continue as a second cohort of small-sized individuals that can be identified as late emergers in the following year. However, very few larvae with black wing pads, and no adults, were collected as late as September or October so it is doubtful if these late larvae can survive at summer temperatures and emerge successfully.

Based on the growth patterns from body length, and head capsule measurements, and from developmental stage analysis, we interpret the life cycle of *C. integrum* as univoltine. The extended emergence and oviposition periods and the winter growth of larvae would place it in the slow, S_1 life cycle pattern of Hynes (1970).

IRONODES NITIDUS. This species is also univoltine, but in contrast to *C. integrum,* the life cycle in Hynes' (1970) classification is a fast, F_1 type. Larvae were absent in August and September. As adults occurred from February to August, the eggs must be in diapause over the summer. Diapause at summer water temperature occurs in *Epeorus pleuralis* (Banks) according to Ide (1935), and Bohle (1972) has demonstrated that decreasing temperature is the cue that breaks diapause in some mayfly eggs.

Practically all larval growth of *I. nitidus* occurred from November through January when the stream was at its coldest (Fig. 3). The progression of size classes and developmental stages is evident during the winter and spring (Figs. 4 & 5), and adult emergence begins when water temperature increases in February or March.

HABITAT PREFERENCE

A total of 4830 mayfly larvae were collected from the three substrates during the year of sampling (Table 1). Of the total mayfly population 43% were collected on wood, 27% from stones, and 30% in kick samples. The high proportion of mayflies on wood debris is probably exceptional. Deposition of fine particles due to the controlled flow of Berry Creek has reduced the habitat available within the stream bed.

C. integrum was the species most closely associated with wood debris at Berry Creek, with 76% of the larvae being found on that substrate (Table 1). *I. nitidus* was well represented on all three substrates, with no marked preference for wood. Though *Baetis* spp. was the most abundant taxon on wood, it was also common in kick samples and on stones so it is not considered to be xylophilous.

The proportions of each developmental stage of *C. integrum* and *I. nitidus* collected from wood substrates are compared in Table 2. About 40% of the stage A larvae of *C. integrum* were collected on rocks or in kick samples but as the larvae matured they showed a very strong preference for wood debris. In contrast, *I. nitidus* showed less of an association with wood as the larvae matured. The data for the mature larvae of *I. nitidus* are at variance with the emergence trap data for adults. Less than 25% of the larvae with black wing pads were found on wood whereas 70% of the adults were collected in traps where wood debris was the dominant substrate.

GUT CONTENT ANALYSIS

The percent composition of gut contents of *C. integrum* and *I. nitidus* larvae is partitioned on a seasonal basis in Table 3. A considerable amount of woody material was evident in both species but it is included as part of the detritus category because of the difficulty of distinguishing some of it from leaf material. Ingestion of wood fragments probably occurs along with the intake of the aufwuchs film rather than as selective feeding on wood. Nevertheless, it is evident that feeding activity does play a part in particle-size reduction of this very refractory material as a result of mechanical breakdown by the scraping type of feeding behavior.

The amount of mineral material observed in guts of both species was surprisingly high, averaging 16% for the full year. Considering the quantity of these fine particles ingested, it is possible that surface-associated microbes are utilized as a nutriment.

Fungal mycelia and a variety of spores were present in large quantities in both species. While fungi would be associated with both leaves and wood, the larvae dissected were only from wood substrates so this is the probable source of the fungi. Mr. Nick Aumen, Department of Fisheries and Wildlife, OSU, has recently informed us that actinomycetes are an important microbial component in wood decay. Actinomycete filaments resemble fungal mycelia in general appearance but the filaments are much finer, with a diameter of <1 μm compared with 5 μm for fungi. Although some portion of our fungal component may be actinomycetes, the appearance of the cell wall, the clamp connections and larger size of mycelia are markers that should mean our fungal component is largely correctly identified.

The percentage of fungi in gut contents of *C. integrum* and *I. nitidus* showed a seasonal periodicity with maximum ingestion (50%) during winter (Table 3). This may be related to the period of greatest fungal growth, or conversely, to the scarcity of diatoms at that time of year. The number of spores in guts was almost equal to the number of mycelia counted. This is further evidence that the larvae are surface scrapers. Small larvae contained the lowest percentage of mycelia which suggests that they were unable to graze efficiently on the mycelial mat or that they selectively use detritus and diatoms as food.

The major difference in gut contents between *C. integrum* and *I. nitidus* and the other mayflies examined was that the former 2 species contained a higher proportion of fungi. *C. integrum* is more of a wood-habitat specialist than *I. nitidus,* as was demonstrated in previous section. Also, the fungal material in *C. integrum* larvae from wood or stones was approximately the same in quality and quantity, whereas *I. nitidus* collected from stones contained more diatoms than those from wood. Thus, the xylophilous habit of *C. integrum* may be associated with fungal food preferences.

Anderson *et al.* (1978) indicated that the life-history strategy of wood borers and gougers frequently entailed long life cycles to compensate for the low food quality of wood substrates. This pattern does not hold for the surface-scraping mayflies; both *C. integrum* and *I. nitidus* are univoltine with similar growth patterns to winter-growing heptageneids that occur on stone substrates.

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Table 1. Proportion of the mayfly population occurring on the three substrate types at Berry Creek, Oregon, July 1979 to June 1980.

	Number Collected	Percent on Each Substrate		
		Wood	Stone	Kick Sample
Cinygma integrum	752	76	15	9
Ironodes nitidus	823	45	31	24
Epeorus (Iron) spp.	555	37	59	4
Cinygmula sp.	428	32	32	36
Rhithrogena sp.	113	21	72	7
Baetis spp.	1413	46	20	34
Paraleptophlebia spp.	324	15	8	77
Ephemerella spp.	388	26	5	69
Ameletus spp.	34	24	32	44
Total Mayflies	4830	43	27	30

Table 2. Total number collected and proportion of each developmental stage of 2 mayflies collected from wood substrates, Berry Creek, Oregon, July 1979 to June 1980.

	Developmental Stage				
	A	B	C	D	E
Cinygma integrum					
No. collected	295	116	126	196	19
% on wood	60	82	89	86	95
Ironodes nitidus					
No. collected	293	76	253	158	43
% on wood	55	46	39	40	23

Table 3. Percentage composition of food types in the guts of *Cinygma integrum* and *Ironodes nitidus*, Berry Creek, Oregon, July 1979 to June 1980.

	No.	Detritus*	Fungi	Diatoms**	Mineral
C. integrum					
June-Aug	30	35	25	18	22
Sept-Nov	42	35	33	6	26
Dec-Feb	34	25	50	13	12
Mar-May	50	65	24	7	4
MEAN		40	33	11	16
I. nitidus					
June-Aug	6	80	12	0	8
Sept-Nov	18	32	26	7	35
Dec-Feb	40	25	60	6	9
Mar-May	40	60	19	11	10
MEAN		49	29	6	15

* Includes wood, leaves, and unidentifiable material.

** Includes filamentous algae.

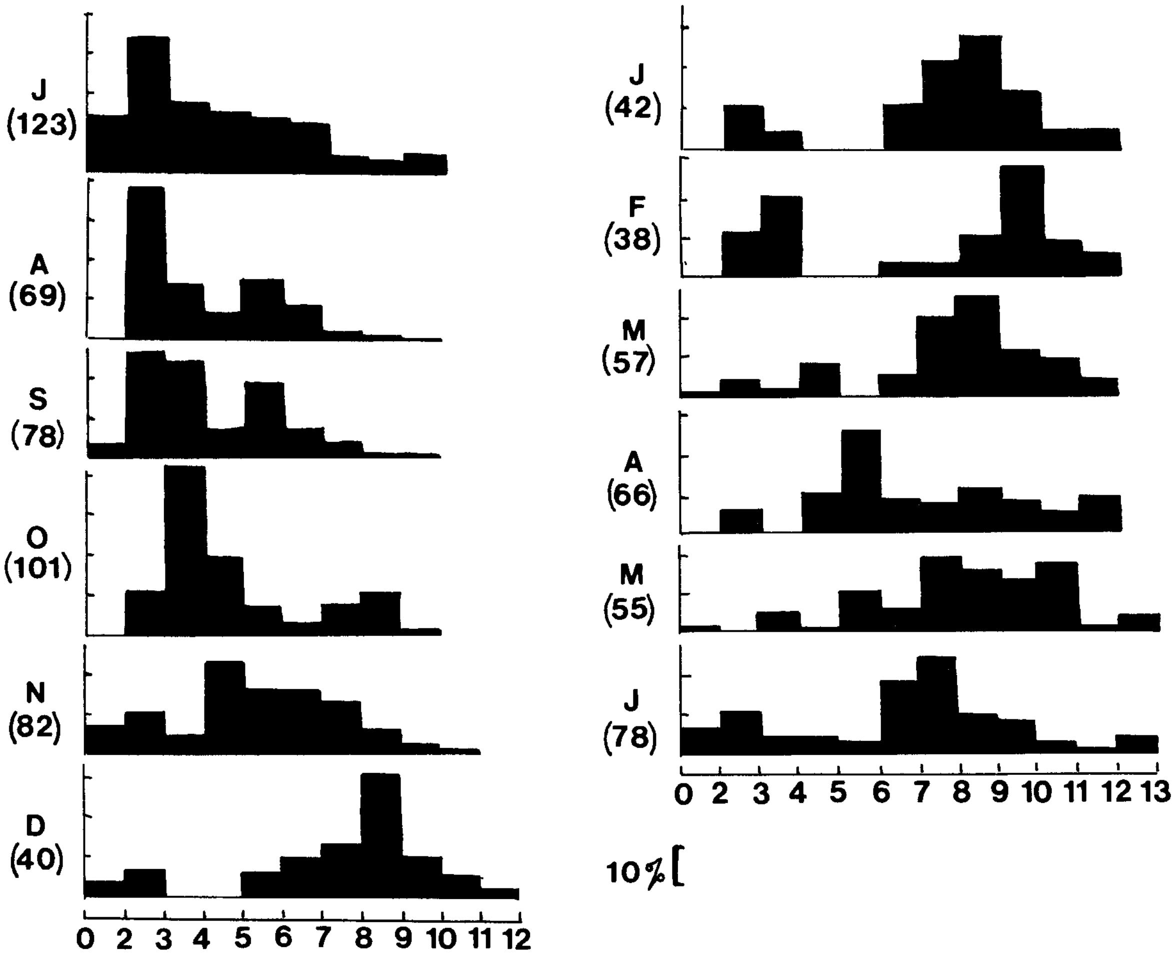

Fig. 1. Percentage composition of size classes (in mm) of *Cinygma integrum* larvae at Berry Creek, July 1979 to June 1980. Numbers in parenthesis indicate sample size.

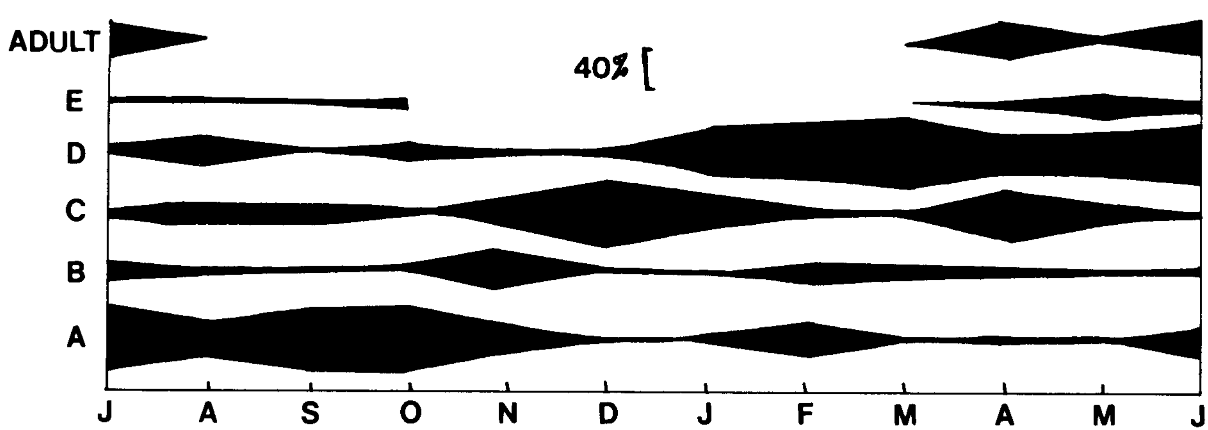

Fig. 2. Percentage composition of the developmental stages of *Cinygma integrum* at Berry Creek, July 1979 to June 1980.

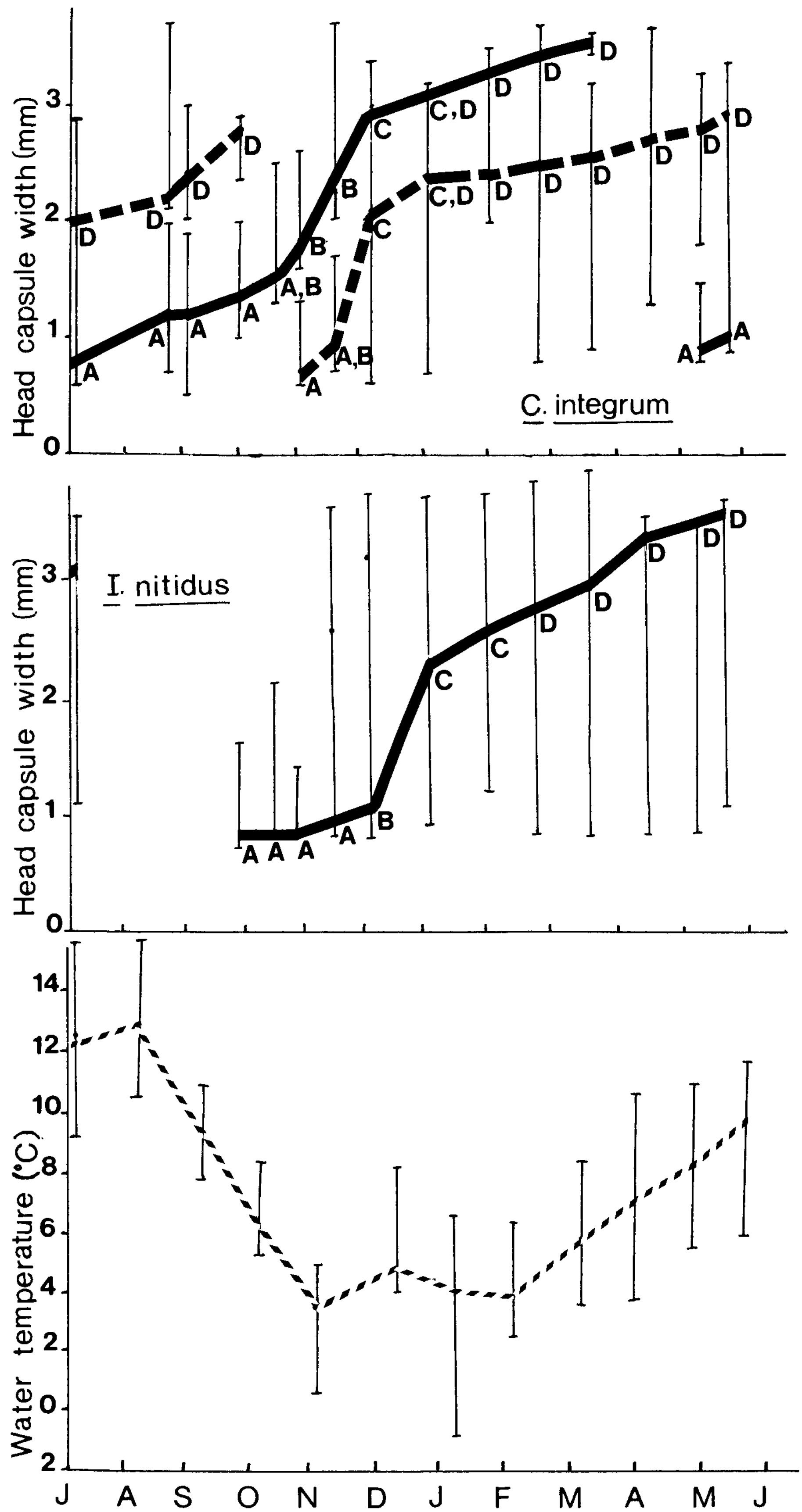

Fig. 3. Range and modal values of head capsule width of *Cinygma integrum* and *Ironodes nitidus* at Berry Creek, July 1979 to June 1980. Predominant developmental stages at each modal point are indicated. Maximum, mean and minimum water temperatures are also given.

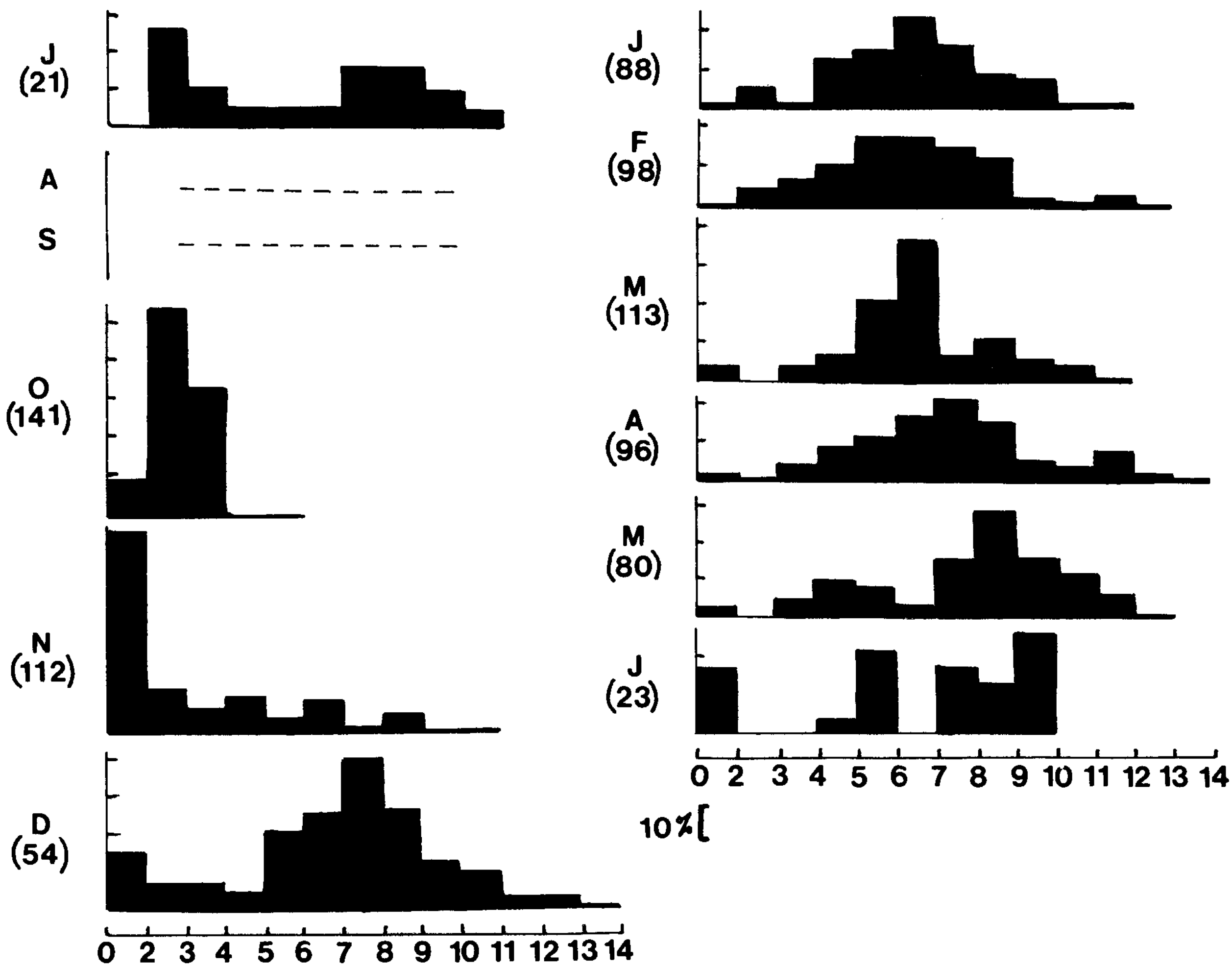

Fig. 4. Percentage composition of size classes (in mm) of *Ironodes nitidus* larvae at Berry Creek, July 1979 to June 1980. Numbers in parenthesis indicate sample size.

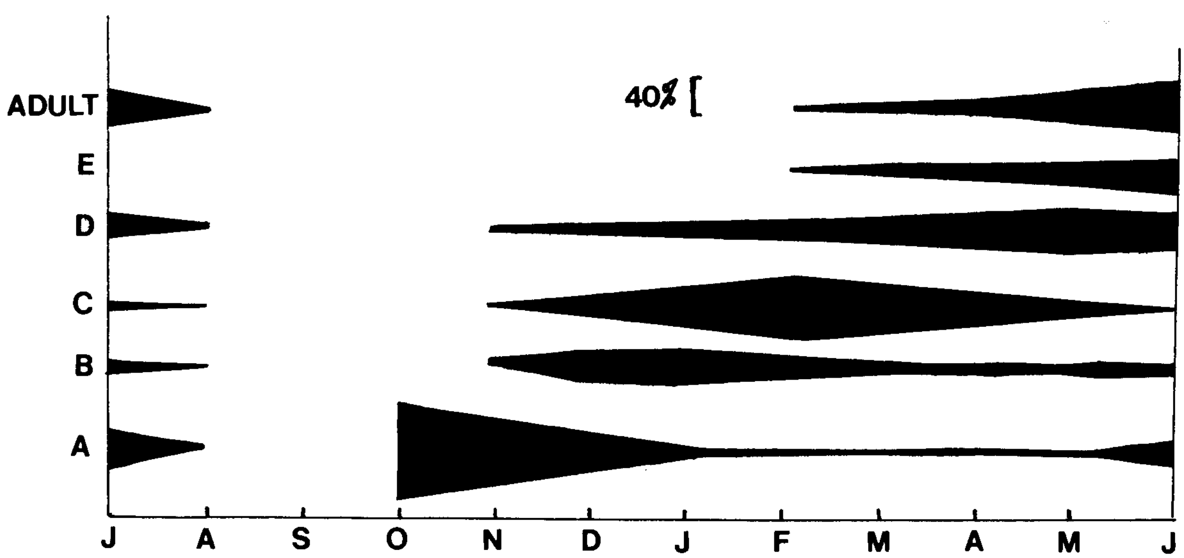

Fig. 5. Percentage composition of the developmental stages of *Ironodes nitidus* at Berry Creek, July 1979 to June 1980.