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*Simulium bernerii* Freeman (Diptera: Simuliidae) and its  
mayfly host (Ephemeroptera: Oligoneuridae)

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SYNOPSIS

An analysis is given of collections of *Elassoneuria* nymphs bearing immature *Simulium bernerii* from the forest zone of West Cameroon near Kumba. The size and sex of the mayfly nymphs, the size and attachment sites of *S. bernerii* and the infestation rates are recorded. The method of dispersion is discussed.

*Simulium bernerii* Freeman (1954) was originally discovered in Togoland (Berner, 1954). It has since been reported from the forest zone of East Cameroon (Grenier *et al.*, 1961) and elsewhere (e.g. Crosskey, 1965). Also a new subspecies, *S. bernerii kumboense* Grenier, Germain & Mouchet (1965a, b) was recorded in the mountain savanna zone of West Cameroon. Crosskey (1965) has summarised what little is known of this species.

*S. bernerii*, of a form differing slightly from the type form, has been found to be common in the forest zone of West Cameroon near Kumba (Lewis *et al.*, 1969). The material on which this paper is based was collected during a survey primarily designed to discover the early stages of *S. dukei* Lewis, Disney & Crosskey, whose larvae and pupae were eventually found on prawns (Disney, 1969). The observations reported represent the accumulated notes on the material of *Elassoneuria* Eaton collected during this survey. The nymphs of *Elassoneuria* are not identifiable to the species level in the present state of knowledge of the African mayfly fauna; adults reared from nymphs collected near Kumba have, however, proved to be an undescribed species. The material has been deposited with Dr M.T. Gillies, who will be preparing a description in due course.

METHODS

The mayfly nymphs were collected by means of hand-nets (standard F.B.A. circular frame nets of 8 meshes/cm). The nymphs were most frequently procured from tangles of dead leaves and debris caught round sticks and stones in fast flowing rivers. Some were found under stones or in trailing root mats.

The rearing of adult mayflies for identification purposes proved to be more difficult than anticipated. No satisfactory procedure was devised, but the following arrangement

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produced a few adults from initially large batches of nymphs. A plastic box was placed in the laboratory sink, small holes having been previously made in one end (about one-third of the height measured from the bottom). The unpierced end was then raised so that when the box contained water the bottom was scarcely covered at the raised end. Into this box was placed a strip of bamboo with its lower end submerged in the deep end and its upper end clear of the water against the raised end of the box. A flow of tap water (untreated water from Barombi Mbo Lake) was then directed down this bamboo strip by means of a rubber tubing siphon. Nymphs were placed in the box with a few dead leaves, and the top was covered over with cotton gauze. The nymphs tended to climb the bamboo strip and aggregate in the shallow flowing water.

Nymphs not used for rearing but collected as part of the survey were examined alive for blackfly pupae. Any pupae were retained for rearing. The wing case bearing the pupa was removed, and its underside cleaned of adhering tissue. The pupa was then rinsed in clean water and placed in a glass tube in the bottom of which was a damp cotton pad. The mouth of the tube was closed with cotton gauze (secured by a cork with a hole in the middle). The tubes of pupae were laid on their sides on a damp cloth in a shallow dish. Pupae were examined regularly and washed once a day until the adult hatched.

After examination for pupae the nymphs were put singly into tubes of alcohol for subsequent detailed examination under a dissecting microscope. An occasional larva became dislodged, so that it was necessary to check the tube for detached larvae when recording infestation rates.

All nymphs were measured to the nearest half millimetre—the length from the front of the head to the tip of the abdomen (excluding the tails) being chosen as a measure of size. The numbers and positions of all black-fly larvae, pupae, cocoons and pupal pelts were recorded, and the lengths of the larvae, to the nearest half millimetre, were measured. It is not yet possible to relate larval lengths to the developmental instars.

#### NOTES ON THE MAYFLIES

*Elassoneuria* only seem to be found in rivers in which free-living *Simulium* are abundant. Apart from nymphs collected for rearing purposes, 902 *Elassoneuria* were collected from rivers near Kumba (these rivers are shown on a sketch map in Disney, 1971). The size distribution of these nymphs is plotted in figure 1 (top). The deficiency of smaller nymphs is striking. Whether this is entirely due to the inadequacy of the collecting technique as a means of sampling the total population, or whether in addition the younger nymphs are normally in different microhabitats from the older nymphs, is not known.

Above 13 mm. in length the male nymphs are readily distinguished from the females by their larger, inflated eyes. Of 670 nymphs over 13 mm. in length collected, only 176 (26.3 per cent.) were males; this ratio of about 3 females to every male is surprising. The females grow larger than males (fig. 1, centre and bottom), and it has been found (from the few successfully reared adults) that male nymphs undergo ecdysis to the adult when they are about 19 mm. long, and females when they are about 24 mm. This difference in size at maturity would go some way towards explaining the differences between the size distribution patterns of males and females (fig. 1) but would be quite inadequate to explain the enormous predominance of female nymphs.

When the different localities were considered separately, it was found that the sex ratio showed wide variations. The Meme river just upstream of the entry of the Bille

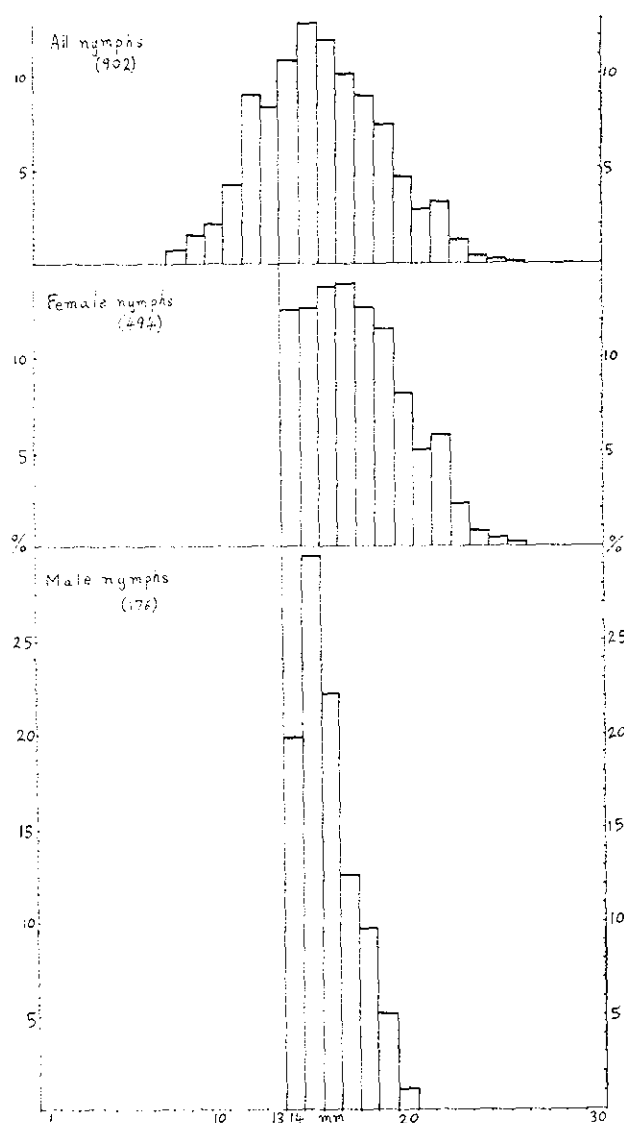


Fig. 1. Size distribution of *Ellassoneuria* nymphs collected near Kumba. Horizontal axis, lengths of nymphs in mm., vertical axis, percentages of nymphs. Top, all nymphs (total 902); centre, female nymphs greater than 13 mm. (total 494); bottom, male nymphs greater than 13 mm. (total 176).

tributary had 6 males in a sample of 66 nymphs greater than 13 mm. in length (9.1 per cent. males). Within 500 m., in the Bille, there were 20 out of 117 (17.1 per cent. males). The Kobe river (a tributary of the Mungo) had 33 males out of 124 nymphs (26.6 per cent. males) where it passes Etam village, but 6.5 km. upstream (by Ikiliwindi village) there were 43 males out of 114 nymphs (37.7 per cent. males). The neighbouring Menge river (North of Ikiliwindi) had 52 males out of 176 nymphs (29.5 per cent. males). This

variation suggests some environmental factor is influencing the sex ratio, but a comparison of several localities at several times of year is required. Anomalies in mayfly sex ratios have been reported elsewhere, together with parthenogenesis (Needham *et al.*, 1935).

When trying to rear adults in a large flask on its side, with a stream of water constantly flushing the flask, a further anomaly was observed. Some of the female nymphs died, but their bodies were not removed and in a few days they had disintegrated, releasing clumps of greenish eggs. The remains of the nymphs disappeared within a few days, but the eggs remained in good condition for several weeks. Another female nymph in the same flask started ovipositing and laid small clusters of eggs on the dead leaves and on the walls of the flask. These eggs, and the clumps released by disintegrating females, remained in good condition for more than a month. The flask was then drained of water and dried out, resulting in the destruction of the eggs. It is therefore possible that parthenogenesis may occur in the Kumba species of *Elassoneuria*, which would help to explain the anomalous sex ratios observed; at least these anomalies merit further investigation. The adults appear to be strictly nocturnal. Eight adult *Elassoneuria* were hatched in the laboratory, all emerging by night, and two adults were caught on the windscreen of the Land Rover when driving at night.

#### NOTES ON *Simulium bernerii*

##### *Infestation rates*

Crosskey (1965), in discussing the limited records available to him, refers to the "infestation rate" of *S. bernerii* on *Elassoneuria* nymphs. In the present study it soon became apparent that the infestation rate (defined here as the mean number of immature *S. bernerii* per *Elassoneuria* nymph) varied not only from locality to locality but within any one locality from size-class to size-class of the nymphs. In Table 1 collections from eight localities are analysed in terms of three size-classes of nymph. The small nymphs measured 8-13 mm. in length, the medium 14-19 mm. and the large nymphs 20-25 mm. Seven rivers are listed: Kumba lies to the western edge of the drainage basin of the River Mungo; to the west of Kumba lies the drainage basin of the River Meme; the Bille river is a tributary stream of the Meme (crossing the Kumba-Mbonge road at Kwakwa); and the other five rivers are tributaries of the Mungo, the Blackwater (crossing the Kumba-Loum road by Ebonje) flowing into the Mungo from the east side and the other four rivers entering from the west side (all crossing the Kumba-Mamfe road, the Kobe also crossing the Kumba-Loum road).

The populations sampled tended to be either rather old (e.g. Menge) or rather young (e.g. Meme, Kobe), so that adequate numbers of nymphs for all three size-classes were not obtained in any one sample. In Table 1 the Meme sample shows clearly that medium nymphs have a higher infestation rate with *S. bernerii* than small nymphs (1.1 and 0.6, respectively), as does the sample from Kobe (Etam) (2.1 and 1.5, respectively). The older population sample from the Menge shows clearly that large nymphs have a higher infestation rate than medium nymphs (1.3 and 0.8, respectively). It seems reasonable to conclude that the infestation rate tends to increase with the size and therefore presumably with the age of the nymphs. The explanation is probably partly that the longer a nymph is available the more *S. bernerii* are likely to find it. It is apparent that any discussion of infestation rates of *S. bernerii* in different localities, or at different times of year in one

Table 1. Infestation rates of *S. berneri* on nymphs of *Elassoneuria* (Kumba District, West Cameroon).

River	Nearest village to collecting point	Months	Small nymphs				Medium nymphs				Large nymphs						
			No. of nymphs	No. of larvae	No. of pupae	Mean no. per nymph	No. of nymphs	No. of larvae	No. of pupae	Mean no. per nymph	No. of nymphs	No. of larvae	No. of pupae	Mean no. per nymph	Total <i>S. berneri</i>	Total no. per nymph	
Meme	Kwakwa	Oct/Nov. 1968	28	17	0	17	0.6	59	62	3	65	1.1	6	8	1	9	1.5
Bille	Kwakwa	Oct./Nov. 1968	71	2	0	2	0.03	100	4	0	4	0.04	5	0	0	0	0
Blackwater	Ebonji	Dec. 1968--Ap. 1969	0	0	0	0	0	17	0	0	0	0	10	0	1	1	0.1
Kobe	Etam	Oct. 1968	57	81	2	83	1.5	105	214	5	219	2.1	13	37	0	37	2.8
Menge	Ikilwindi	Oct. 1968	38	24	0	24	0.6	106	59	1	60	0.6	7	3	0	3	0.4
Wowe	Ikilwindi	Nov. 1968	7	6	0	6	1.0	114	88	1	89	0.8	55	58	11	69	1.3
Dilolo	Baduma	Apr. 1969	3	0	0	0	0	7	1	0	1	0.1	8	1	0	1	0.1
	Bolo	Oct. 1968	2	0	0	0	0	20	1	0	1	0.05	9	0	0	0	0

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Table 3. Attachment sites of immature *S. berneri* to medium nymphs of *Elatoneuria*

Attachment sites	Larval length (in mm.)															Totals	
	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	Pupae	Cocoons	(larvae)	Percentage	
Mouth parts	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	1	0.2
Maxillary gills	—	23	14	3	2	—	—	—	—	—	—	—	—	—	—	42	9.3
Head angles	—	—	—	—	1	1	2	4	4	16	6	2	—	—	—	36	7.9
Wing covers	—	—	1	1	2	1	1	—	—	2	—	—	10	13	8	1.8	
Thorax (dorsal side)	—	—	—	1	—	1	—	—	—	—	—	—	—	—	2	0.4	
Coxae + Trochanters	—	3	—	1	3	3	—	—	—	—	—	—	—	—	10	2.2	
Prosternum	—	—	—	9	8	15	8	2	1	3	—	—	—	—	54	11.9	
Mesosternum	—	2	5	5	11	7	3	2	2	4	—	—	—	—	41	9.0	
Metasternum	—	1	—	3	7	3	1	1	—	—	—	—	—	—	17	3.7	
Abdominal Tergites	—	—	3	1	1	—	—	—	—	—	—	—	—	—	5	1.1	
Sternites	—	—	2	1	1	1	—	—	—	—	—	—	—	—	5	1.1	
Gills 1	—	5	4	5	7	2	—	—	—	—	—	—	—	—	23	5.1	
Gills 2	2	17	12	4	—	—	—	—	—	—	—	—	—	—	35	7.7	
Gills 3	1	12	18	8	3	—	—	—	—	—	—	—	—	—	42	9.3	
Gills 4	—	11	19	7	1	—	—	—	—	—	—	—	—	—	38	8.4	
Gills 5	—	10	16	4	—	1	—	—	—	—	—	—	—	—	31	6.8	
Gills 6	1	14	16	8	1	—	—	—	—	—	—	—	—	—	40	8.8	
Gills 7	1	10	8	4	1	—	—	—	—	—	—	—	—	—	24	5.3	





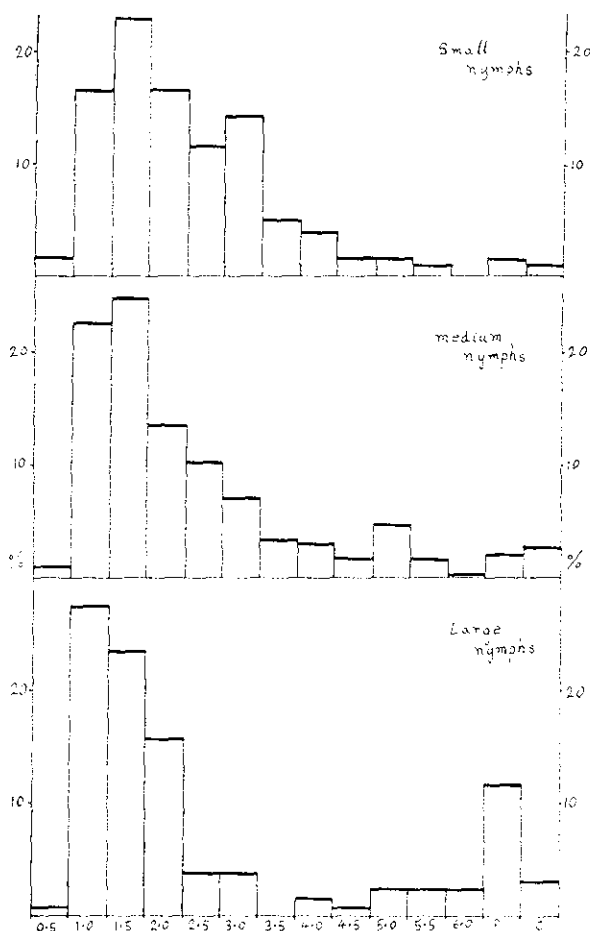


Fig. 2. Size distribution of immature *S. bernerii* on small, medium and large nymphs of *Ellassoneuria*. Vertical axis, percentages; horizontal axis, size classes of larvae in mm. (P, pupae; C, empty cocoons).

locality, must take into account the sizes of the *Ellassoneuria* nymphs. It would be most convenient to use medium nymphs as a standard for comparing different samples. A striking difference in Table 1 is the infestation rate on medium nymphs in October 1968, which was 0.6 in the Kobe river by Ikiliwindi but 2.1 in the same river by Etam village (about 6.5 km. downstream).

#### *Attachment sites and size-distribution of larvae*

Crosskey (1965) listed the attachment sites of larvae of *S. bernerii* to three *Ellassoneuria* nymphs. In the present study, the larger numbers available have shown that the choice of attachment site is influenced primarily by the size of the larva and secondly by the size of the *Ellassoneuria* nymph. Tables 2-4 present the data on attachment sites for 170 larvae, 3 pupae and 2 cocoons recorded on small nymphs, 454 larvae, 10 pupae and 13

cocoons on medium nymphs, and 180 larvae, 15 pupae and 4 cocoons on large nymphs. Figure 2 shows the size distributions of immature *S.berneri* recorded on the three sizes of nymphs. Table 5 summarises the attachment site data. The data indicate that the smallest larvae found on nymphs are first instar larvae (the 0.5 mm. larvae each had an egg-tooth present). This would appear to be the first record of first instar larvae of mayfly-phoretic *Simulium*.

It would appear that it is the first instar larva that has the task of finding its host. The larvae above 0.5 mm. in size found on large nymphs show a population age-structure with a characteristic skew to the left side of the plot (fig. 2, bottom). When the populations on the medium and small nymphs are compared (fig. 2, centre and top) there is seen to be a marked deficiency of 1.0 mm. larvae, especially on the small nymphs. In addition, the tables show that young larvae on large nymphs characteristically settle on the gills, older larvae move on to the thorax, and the oldest larvae are on the head and wing covers. However, the smaller the nymphs the higher the proportion of young larvae that settle on the thoracic sterna and elsewhere. Thus the mean larva size of 3.2 mm. on the thoracic sterna of large nymphs (Table 5) is lowered by a dilution of

**Table 5.** Summary of attachment sites of immature *Simulium berneri* to small, medium and large nymphs of *Elassoneuria*.

Attachment sites	Larval length (in mm.)			Pupae + cocoons	Mean size of larvae
	0.5-1.5	2.0-3.0	3.5-6.0		
<i>Small nymphs</i>					
Gills	55	12	1	0	1.4
Thoracic sterna	10	35	9	0	2.6
Head angles	0	7	12	0	} 3.5
Wing covers	0	3	1	5	
Totals	65	57	23	5	
<i>Medium nymphs</i>					
Gills	214	61	0	0	1.4
Thoracic sterna	8	68	36	0	2.9
Head angles	0	2	34	0	} 4.8
Wing covers	1	4	3	23	
Totals	223	135	73	23	
<i>Large nymphs</i>					
Gills	64	18	0	0	1.4
Thoracic sterna	2	8	6	0	3.2
Head angles	0	1	3	0	} 4.6
Wing covers	0	1	1	19	
Totals	66	28	10	19	

small larvae to 2.6 mm. on small nymphs. It seems probable that two factors are operating. First, the older nymphs have been exposed to *S.berneri* for longer; consequently they carry more older larvae and pupae. Secondly, it seems that young larvae experience greater difficulty in trying to stay attached to the smaller gills of small nymphs. Probably the deficiency of 1.0 mm. larvae on small nymphs is due to a higher rate of dislodgement than is experienced on larger nymphs.

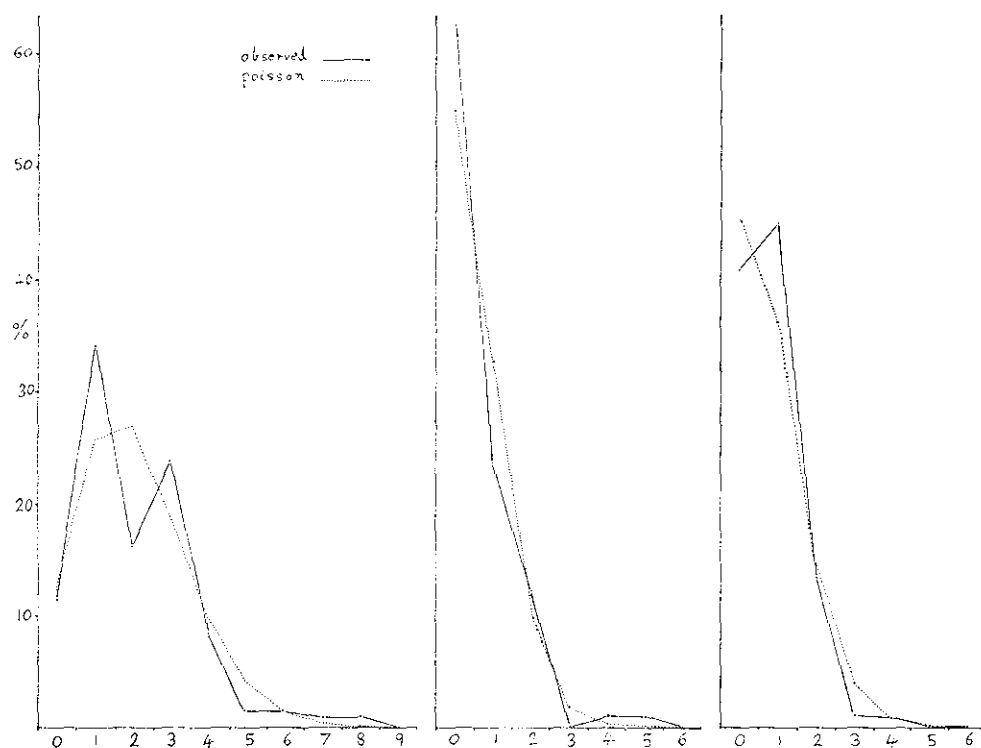


Fig. 3. Dispersion of immature *S.bernerii* on medium-sized nymphs of *Elassoneuria*. Left, Kobe River (Etam), October, 1968; centre, Kobe River (Ikiliwindi), October, 1968; right, Menge River, November 1968. Vertical axis, percentages; horizontal axis, nymphs with 0, 1, 2, etc. *S.bernerii* attached. ————observed, ..... Poisson distribution.

#### Dispersion

Although no experimental evidence of freely drifting small larvae has been obtained, it would appear that the larvae of *S.bernerii* arrive on the mayfly nymphs as very young larvae (and not as eggs; no *Simulium* eggs have been found attached to *Elassoneuria* nymphs). If these young larvae were to hatch from eggs laid on a trailing leaf (or similar substratum), and each one independently was to set off drifting downstream endeavouring to locate an *Elassoneuria* nymph, then one would expect two features to emerge from an analysis of the populations on nymphs. First, there would be an enormous loss of first instar larvae, assuming that the mortality of larvae already established on nymphs is significantly less than among unestablished (still drifting) larvae, and one would expect to find that on the nymphs first instar larvae were relatively scarce compared to the immediately subsequent instars. The data in figure 2 tend to support this prediction.

Secondly, one would expect the dispersion of larvae on nymphs to be random, if they are arriving as single drifting larvae. Compare for example the random dispersion of the larvae of codling moths (*Cydia pomonella* (L.)) in apples, which derives from the female's habit of laying only one egg at a time after locating an apple by chance encounter during randomly directed flight activity (Clark *et al.*, 1967). An *Elassoneuria* nymph constitutes

a discrete natural habitat unit from the point of view of *S.berneri* larvae, so, if they arrive singly and as a result of chance encounter, their dispersion pattern should be random also. The dispersion data for immature *S.berneri* on *Elassoneuria* nymphs has been analysed for the medium-size nymphs from Kobe (Etam), Kobe (Ikiliwindi), and Menge (data in Table 1) and plotted in figure 3: superimposed on each plot is the curve of Poisson distribution calculated from the infestation rate. The close coincidence of the observed dispersions with the Poisson dispersions suggests that *S.berneri* is indeed randomly distributed on *Elassoneuria* nymphs.

The largest number of *S.berneri* larvae per nymph that has been recorded near Kumba was 10 larvae on a medium-sized nymph collected at Kobe river (Etam) on 6th December, 1968. Crosskey (1965) records an *Elassoneuria* nymph from Uganda which was carrying 11 larvae and 1 pupa of *S.berneri*. This suggests that the infestation rate of the population sampled was probably at least 2.0 on medium-sized nymphs (assuming a random dispersion).

#### Laboratory reared adults

Seventeen *S.berneri* (12 females and 5 males) were successfully reared from pupae in the laboratory. Five of these emerged by night, and the rest by day.

#### SUMMARY

*Simulium berneri* Freeman living in association with the nymphs of an undescribed species of *Elassoneuria* is common near Kumba in the forest zone of West Cameroon. First instar larvae of *S.berneri* have been recorded for the first time.

Males and females of *Elassoneuria* show somewhat different patterns of size distribution. Females attain a greater size than males before eclosion of the adult. The sex ratio of the nymphs varies from locality to locality, but overall there are about 3 females to every male. Adult eclosion takes place by night.

The infestation rates of *S.berneri* on *Elassoneuria* nymphs at eight localities varied with the locality and also with the size of the nymphs within any one locality. The choice of attachment site varies with the size of the *Simulium* larva and the size of the *Elassoneuria* nymph: young larvae apparently experience difficulty in attaching to small nymphs. It is suggested that it is the first instar larvae which locate the mayfly host. Analysis of the dispersion data for *S.berneri* on the nymphs suggests that they are randomly distributed on the nymph population; this supports the notion that the young larvae arrive on their hosts by drifting and chance encounter.

The following members of the Unit's staff helped collect mayfly nymphs: Mr R.E.Oguama, A.Alim, A.Iburu, O.Mbeng, F.Moh, and E.Ndifon.

Dr M.T.Gillies (University of Sussex) kindly examined the mayfly material and also advised on mayfly literature.

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